Documentation related to Session 3 of the meeting
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Malaria Policy Advisory Group (MPAG) Meeting

13 – 15 April 2021 *(CEST time zone)*
Virtual meeting

PROVISIONAL PROGRAMME*

Tuesday, 13 April 2021			
	Session 1	**Open**	
12:00 – 12:05	Welcome by the ADG, UCN	Dr Ren Minghui	
12:05 – 12:15	Welcome by the Chair, MPAG	Dr Dyann Wirth	
12:15 – 13:00	Report from the Director, GMP	Dr Pedro Alonso	
13:00 – 13:30	Partner Perspective, US President's Malaria Initiative	Dr Raj Panjabi	
13:30 – 14:00	Rethinking malaria	Dr Rose Leke & Dr Alastair Robb	**For guidance**
14:00 – 14:15	*Coffee break*		
	Session 2	**Open**	
14:15 – 15:00	Clinical malaria – parasite density thresholds in different transmission settings and implications for use of RDTs	Dr Jane Cunningham	**For guidance**
15:00 – 15:30	Update on the situation of antimalarial drug efficacy and resistance in Africa	Dr Pascal Ringwald	
15:30 – 16:00	Proposed technical consultation to stage *P. knowlesi* along the continuum between zoonosis and human pathogen	Dr Kim Lindblade	
16:00	*End of day*		

Wednesday, 14 April 2021			
	Session 3	**Open**	
12:00 – 12:45	HRP2 gene deletions – a focus on horn of Africa region: Background / Presentation	Dr Jane Cunningham	**For decision**
12:45 – 13:30	Proposed technical consultation on urban malaria: Background /Presentation	Dr Abdisalan Noor	**For guidance**
13:30 – 13:45	*Coffee break*		
	Session 4	**Open**	
13:45 – 14:15	Update on guidance for severe malaria	Dr Peter Olumese	**For decision**

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14:15 – 14:45	Update on the classification of insecticide-treated net products – annual update as requested by MPAG	Dr Jan Kolaczinski & Dr Marion Law	**For guidance**
14:45 – 15:15	Update on digital solutions for malaria elimination surveillance	Dr Abdisalan Noor & Ms Mwalenga Nghipumbwa	
15:15	*End of day*		

Thursday, 15 April 2021			
	Session 5	**Closed**	
12:00 – 15:00	Finalization of wording of recommendations	Dr Dyann Wirth	**For guidance**

** Provisional programme and may be subject to change*

Malaria Policy Advisory Group Meeting

13—15 April 2021, Geneva, Switzerland
Background document for Session 3

HRP2 gene deletions – a focus on horn of Africa region

Dr Jane Cunningham, WHO Global Malaria Programme, Geneva, Switzerland

Prevalence of *Plasmodium falciparum* lacking histidine-rich proteins 2 and 3: a systematic review. Rebecca Thomson et al. Bull World Health Organ 2020;98:558–568F doi: http://dx.doi.org/10.2471/BLT.20.250621

Prevalence of *Plasmodium falciparum* lacking histidine-rich proteins 2 and 3: a systematic review

Rebecca Thomson,[a] Jonathan B Parr,[b] Qin Cheng,[c] Stella Chenet,[d] Mark Perkins[e] & Jane Cunningham[f]

Objective To calculate prevalence estimates and evaluate the quality of studies reporting *Plasmodium falciparum* lacking histidine-rich proteins 2 and 3, to inform an international response plan.

Methods We searched five online databases, without language restriction, for articles reporting original data on *Plasmodium falciparum*-infected patients with deletions of the *pfhrp2* and/or *pfhrp3* genes (*pfhrp2/3*). We calculated prevalence estimates of *pfhrp2/3* deletions and mapped the data by country. The denominator was all *P. falciparum*-positive samples testing positive by microscopy and confirmed positive by species-specific polymerase chain reaction testing (PCR). If microscopy was not performed, we used the number of samples based on a different diagnostic method or PCR alone. We scored studies for risk of bias and the quality of laboratory methods using a standardized scoring system.

Findings A total of 38 articles reporting 55 studies from 32 countries and one territory worldwide were included in the review. We found considerable heterogeneity in the populations studied, methods used and estimated prevalence of *P. falciparum* parasites with *pfhrp2/3* deletions. The derived prevalence of *pfhrp2* deletions ranged from 0% to 100%, including focal areas in South America and Africa. Only three studies (5%) fulfilled all seven criteria for study quality.

Conclusion The lack of representative surveys or consistency in study design impairs evaluations of the risk of false-negative results in malaria diagnosis due to *pfhrp2/3* deletions. Accurate mapping and strengthened monitoring of the prevalence of *pfhrp2/3* deletions is needed, along with harmonized methods that facilitate comparisons across studies.

Abstracts in عربي, 中文, Français, Русский and Español at the end of each article.

Introduction

Despite improvements in malaria control over the past decade, malaria caused an estimated 405 000 deaths worldwide in 2018.[1] In 2010, World Health Organization (WHO) treatment guidelines established that all cases of suspected malaria should be confirmed by microscopy or an antigen-detecting rapid diagnostic test before treatment.[2] Malaria rapid diagnostic tests contain one or a combination of antibodies that recognize specific plasmodial antigens. These antigens include histidine-rich protein 2 (HRP2) which is specific to *P. falciparum*, and genus- and species-specific lactate dehydrogenase or aldolase, which are produced by all four major human-infecting *Plasmodium* species.[3] The number of rapid diagnostic tests procured has increased significantly, from 10 million in 2002 to 412 million in 2018.[1] The great majority of these tests detect an HRP2 target, alone or with another antigen, with 15 of 16 (94%) WHO-prequalified malaria tests targeting HRP2 for *P. falciparum* detection.[4]

Rapid diagnostic tests targeting HRP2 came to dominate the market because they are generally more sensitive than other assays and tend to be more heat stable.[5,6] The presence of repetitive epitopes in HRP2 provides numerous antibody binding sites and enables the detection of low levels of protein. The monoclonal antibodies used in HRP2-detecting tests often cross-react with HRP3, encoded by the *pfhrp3* gene,[7,8] particularly at parasite counts above 1000 per µL of blood.[9] HRP3 is a structural homologue of HRP2 that shares similar amino-acid repeats.[8,10]

Deletions in the *pfhrp2* and/or *pfhrp3* (*pfhrp2/3*) genes as a cause of false-negative rapid diagnostic tests was first recognized in 2010 in the Peruvian Amazon basin.[11] Molecular testing by polymerase chain reaction (PCR) confirmed *P. falciparum* infection, but also that *pfhrp2* and *pfhrp3* genes were deleted in 41% (61 samples) and 70% (103 samples) of these 148 samples, respectively.[11] Additional analyses have confirmed a significant increase in the frequency of samples showing *pfhrp2/3* deletions in the same area.[12,13] More recently, malaria parasites with *pfhrp2/3* gene deletions have been documented in other parts of the world including East,[9,14] Central,[15] West[16] and Southern Africa,[17] Asia[18] and the Middle East.[19] Most concerning was a study in Eritrea that reported samples from 62% (31/50) of microscopy-confirmed *P. falciparum* patients testing negative for *pfhrp2*.[20] Collectively, these reports suggest a global threat to the continued use of HRP2-based rapid diagnostic tests.

In 2014, recommendations on investigating and accurate reporting of *pfhrp2/3* gene deletions were published.[21] Additional criteria have been proposed in more recent studies, including parasite quantification by microscopy or quantitative PCR to rule out false-negative *pfhrp2* detection in samples below the limit of detection of the *pfhrp2* assay,[9] and analysis of *pfhrp3*.[22] However, we have found no assessments of the uptake of these recommendations.

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There are increasing numbers of reports documenting the threat of mutant parasite genes for malaria case management. However, due to different study designs and laboratory methods it is difficult to compare findings across studies and accurately understand this threat. We aimed to compile all published studies on the prevalence of *pfhrp2/3* gene deletions and assess the quality of methods and reporting. We used our findings to paint a global picture of the current status of *pfhrp2/3* deletions to guide decisions on the locations and methods of future surveys.

Methods

Search strategy and data extraction

We carried out a systematic review according to the Preferred Reporting Items for Systematic Reviews and Meta-analyses statement.[23] We made a search of the online databases of PubMed®, Scopus, LILACS (Literatura Latino-Americana e do Caribe em Ciências da Saúde), WHO Global Index Medicus and the Web of Science for articles published in any language between 1 January 2010 and 20 August 2019. We used the search terms "[histidine* OR hrp* OR pfhrp*] AND [deletion* OR variation OR diversity OR lack] AND [malaria OR falciparum]" to identify articles reporting molecular analysis of *P. falciparum* parasite samples for *pfhrp2/3* deletions. Additional articles were identified through manual searches. Further information about the search criteria are provided in Table 1. Two investigators screened the titles and abstracts of all eligible articles and extracted the following information from the full text: country, study sites, study design, year(s) of data collection, patient symptom status, age range, number of *P. falciparum*-positive patients, type of blood sample, which samples underwent molecular analysis, number of samples with *pfhrp2/3* deletions, laboratory methods (seven items; Box 1) and analysis of flanking genes. Discrepancies in the data were double-checked.

Prevalence estimates

To maximize consistency in calculating prevalence across studies, we used the total number of *P. falciparum* samples testing positive by microscopy and confirmed *P. falciparum*-positive by species-specific PCR as the denominator. We did this regardless of whether all or only a subset of patient samples were tested for *pfhrp2/3* deletion by molecular analysis or whether it was the denominator reported in the original publication. If microscopy was not performed, we used the number of samples based on a different diagnostic method or PCR alone. We did not make prevalence estimates from case reports. All prevalence estimates in this review were therefore derived using a standardized denominator and not necessarily the same prevalence as reported in the original article.

Where researchers collected samples from multiple countries, or used different sampling methods or time frames, we separated the results by country or sample collection group to present prevalence data as separate studies. We presented compiled results for studies which collected samples at one point in time from multiple sites with the same sampling design. When we combined data from different studies by country,

Table 1. **Inclusion and exclusion criteria for selection of studies in the systematic review of *Plasmodium falciparum pfhrp2/3* gene deletions**

Characteristic	Included	Excluded
Study population	All ages and populations	None
Study outcome	Percentage of samples testing negative for *pfhrp2* gene, with or without analysis of *pfhrp3* gene	Studies which analysed variation in *pfhrp2* genetic sequence only
Method of confirmation of *pfhrp2* and/or *pfhrp3* gene deletion	Molecular analysis of *pfhrp2/3* gene deletions	Suspected deletions based on rapid diagnostic testing, microscopy or serological testing only
Study design	All, including case studies, cross-sectional or convenience studies	None
Type of paper	Published articles of an original study	Review articles, doctoral theses, abstracts with no corresponding published article
Patient status	Symptomatic suspected malaria patients or asymptomatic people	None
Area of data collection	All countries and regions	None
Date of study publication	1 January 2010 to 20 August 2019	Prior to 1 January 2010

Box 1. **Assessment of study quality in the systematic review of *Plasmodium falciparum pfhrp2/3* gene deletions**

We assessed a total of seven criteria for quality of laboratory methods, five based on recommendations from previous reasearch[21] and two additional criteria.[9,22] The number and percentage of studies complying with each quality criterion were as follows ($n = 55$ studies):

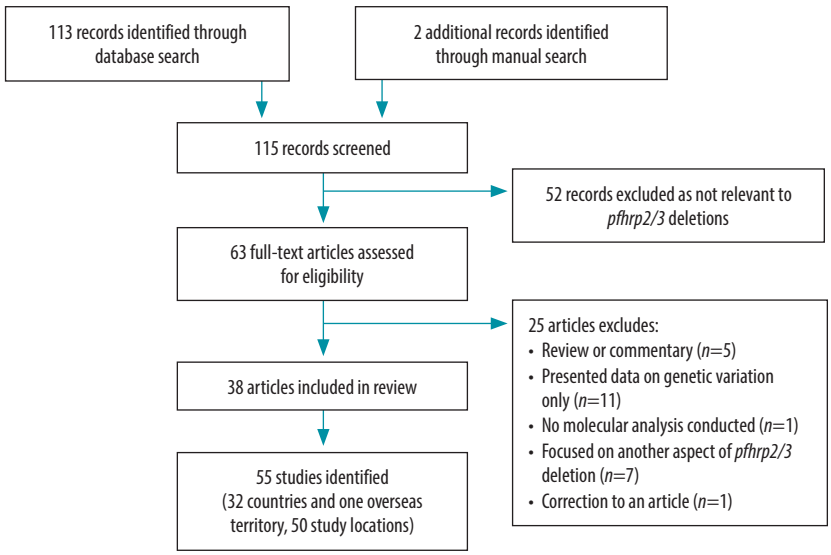
- Quality-assured microscopy: 45 studies (82%).
- *Plasmodium falciparum* species confirmation by PCR test: 55 studies (100%).
- Detection of two other single-copy genes: 21 studies (38%).
- HRP2 detection by serological analysis or using a second brand of WHO-prequalified HRP2-detecting rapid diagnostic test: 13 studies (24%).
- Detection of *pfhrp3* gene by PCR test: 46 studies (84%).
- Use of WHO-prequalified rapid diagnostic test: 27 studies (49%).
- Parasite density quantification: 36 studies (66%).

We awarded one point per criterion satisfied and assigned a total quality score for each study (from 1 to 7), as follows:

Score 1: 0 studies (0%); Score 2: 6 studies (11%); Score 3: 5 studies (9%); Score 4: 16 studies (30%); Score 5: 15 studies (27%); Score 6: 10 studies (18%); Score 7: 3 studies (6%).

HRP2: histidine-rich protein 2; PCR: polymerase chain reaction; WHO: World Health Organization.

Fig. 1. **Flowchart of the literature search in the systematic review of *Plasmodium falciparum pfhrp2/3* gene deletions**



Note: One article reported samples collected from three countries but did not present the results separately, so it is presented as one study.

we weighted the percentage of samples with *pfhrp2* gene deletions to account for differing sample sizes. We used the middle year of the data collection period for studies spanning multiple years.

Assessment of study quality and bias

We assigned a total quality score from 1 to 7 to each study, based on fulfilment of seven criteria for quality of laboratory methods (Box 1).

We assessed study bias as a score from 1 (lowest bias) to 4 (highest bias). The values show the potential bias of the derived prevalence estimate from the true prevalence in the population, depending on the sample population (symptomatic, asymptomatic, mixed or unrepresentative) and samples tested for *pfhpr2/3* genes (all, discordant only or another subset). Studies which analysed all samples have a lower bias score than those which only analysed discordant or a subset of samples, while studies which included both symptomatic and asymptomatic samples have a lower study bias than those which only analysed samples from symptomatic people or an unrepresentative sample.

Results

After screening 115 articles, we included 38 articles in the review (Fig. 1).[11–20,24–50] Within the articles we identified 55 distinct studies conducted in 32 countries and one territory in the regions of Africa, Americas, South-East Asia and Eastern Mediterranean (Table 2; available at: http://www.who.int/bulletin/volumes/98/8/20-250621).

Study characteristics

The included studies showed substantial differences in study design, laboratory methods and data reporting.

Sample populations

The number of samples tested for *pfhrp2* ranged from 1 to 783, while the denominator of *P. falciparum*-positive samples ranged from 1 to 3291 (Table 2). Out of the 55 studies, 36 (65%) analysed blood samples only from people with symptoms of malaria, as part of a prospective or retrospective survey including unbiased cohorts. Samples in these studies were collected from suspected malaria patients presenting to health facilities or through active case detection. Eight studies (15%) included samples from asymptomatic and symptomatic people as part of cross-sectional surveys or malaria screening programmes, while eight other studies (15%) used samples from an unrepresentative sample of participants and three studies (5%) did not specify the symptom status of the participants. One study collected samples from patients with severe malaria only, while one study collected equal numbers of samples from human immunodeficiency virus-positive and -negative children.

In 35 studies (64%) all samples underwent *pfhrp2/3* genotyping. Thirteen studies (24%) genotyped discordant samples only. Of these, nine studies analysed only microscopy-positive and HRP2-rapid diagnostic test-negative samples (of which two were case studies including only one sample), while four studies genotyped only samples which were negative by HRP2-rapid diagnostic test and positive by PCR. One article reporting seven studies only genotyped samples showing the lowest HRP2 concentrations by enzyme-linked immunosorbent assay.

Study procedures

Only three studies (5%) fulfilled all seven criteria for quality of procedures (Box 1). While the number of *P. falciparum*-positive samples was based on microscopy- and PCR-positive results in 45 studies (82%), in nine studies (16%) the denominator was based on PCR results alone, and in one study (2%) it was based on *P. falciparum*-specific lactate dehydrogenase-based rapid diagnostic tests and confirmed by PCR. The presence of *P. falciparum* was confirmed most commonly by amplification of the multi-copy *18SrRNA* gene. Thirty-four studies (62%) analysed samples from dried blood spots, 13 (24%) used venous blood, seven (13%) used a combination of both and one study (2%) did not provide information on sample type. Forty-six studies (84%) conducted molecular analysis to determine *pfhrp3* deletion. One of these studies only genotyped *pfhrp3* deletions among samples found to be *pfhrp2*-negative.

Twenty-one studies (38%) did not amplify any other single-copy genes while 13 studies (24%) amplified one other and 21 studies (38%) amplified at least two other single-copy genes. To rule out negative *pfhrp2/3* PCR results being due to parasite density below the limit of detection of the assay, only samples which were positive by other single-copy genes and failed to amplify the *pfhrp2/3* gene were considered to be *pfhrp2/3*-deleted in the 21 studies which conducted this analysis. The most commonly selected genes for confirmation were *P. falciparum* merozoite surface proteins 1 and 2, and glutamate-rich protein. One study confirmed *pfhrp2*

deletion by testing for *pfhrp3*. However, while parasite density was measured in 36 studies, only five studies used these results when determining if a sample was *pfhrp2/3*-negative. In three studies only samples above a chosen parasite density or deoxyribonucleic acid (DNA) concentration were tested for *pfhrp2*, while in one study all samples below 5 parasites per μL of blood were discounted and in one study samples were only included in the original study if they were above 2000 parasites per μL of blood.

Twenty-nine studies (53%) amplified both exons 1 and 2 of the *pfhrp2* gene, while 26 studies (47%) amplified only exon 2 and 28 studies (51%) amplified the flanking genes of *pfhrp2*. The studies that amplified exon 1 were not necessarily those that amplified flanking genes, with 19 studies (35%) amplifying both exon1 and flanking genes.

Prevalence estimates

The derived prevalence of *pfhrp2* gene deletions in the 55 studies ranged from 0% to 100% (Table 2). Although we present the overall results by study, 14 studies were conducted over many sites and showed geographically heterogenous results. Further details about the results presented by region are provided in the data repository.[51]

In Fig. 2 we mapped the geographical distribution of the highest derived prevalence estimate of *pfhrp2* gene deletions by study for each country. The highest derived prevalence was above 50% in Colombia, Eritrea and Peru. Fig. 3 plots the weighted average prevalence of *pfhrp2* gene deletions for each country and the range by study sites. The weighted average prevalence ranged from 0% to 43%. Average prevalence above 20% was found in Eritrea, Ghana, Nicaragua, Peru and Sudan.

We plotted the prevalence of *pfhrp2*/3 gene deletions by sample size (available in the data repository).[52] Five studies had a sample size over 1000, while 36 had sample sizes smaller than 100. All seven studies reporting greater than 50% prevalence of *pfhrp2* deletions had a sample size of fewer than 55. Scatter plots of prevalence against time are available in the data repository.[53]

Risk of bias

Table 2 shows the bias scores of the prevalence estimates from the true prevalence of *pfhrp2/3* gene deletions in the population. Six studies (11%) had a bias score of four while two (4%) had a bias score of one.

Discussion

We found that mutant parasites have been reported from all major malaria-endemic areas, in asymptomatic and symptomatic *P. falciparum*-positive patients. However, our results also confirm that the full extent of the threat has not yet been characterized. The limited number of well conducted prevalence surveys in malaria-endemic countries indicate geographical variability in the prevalence of mutations in the *pfhrp2* and *pfhrp3* genes and do not completely illuminate the factors driving these differences.

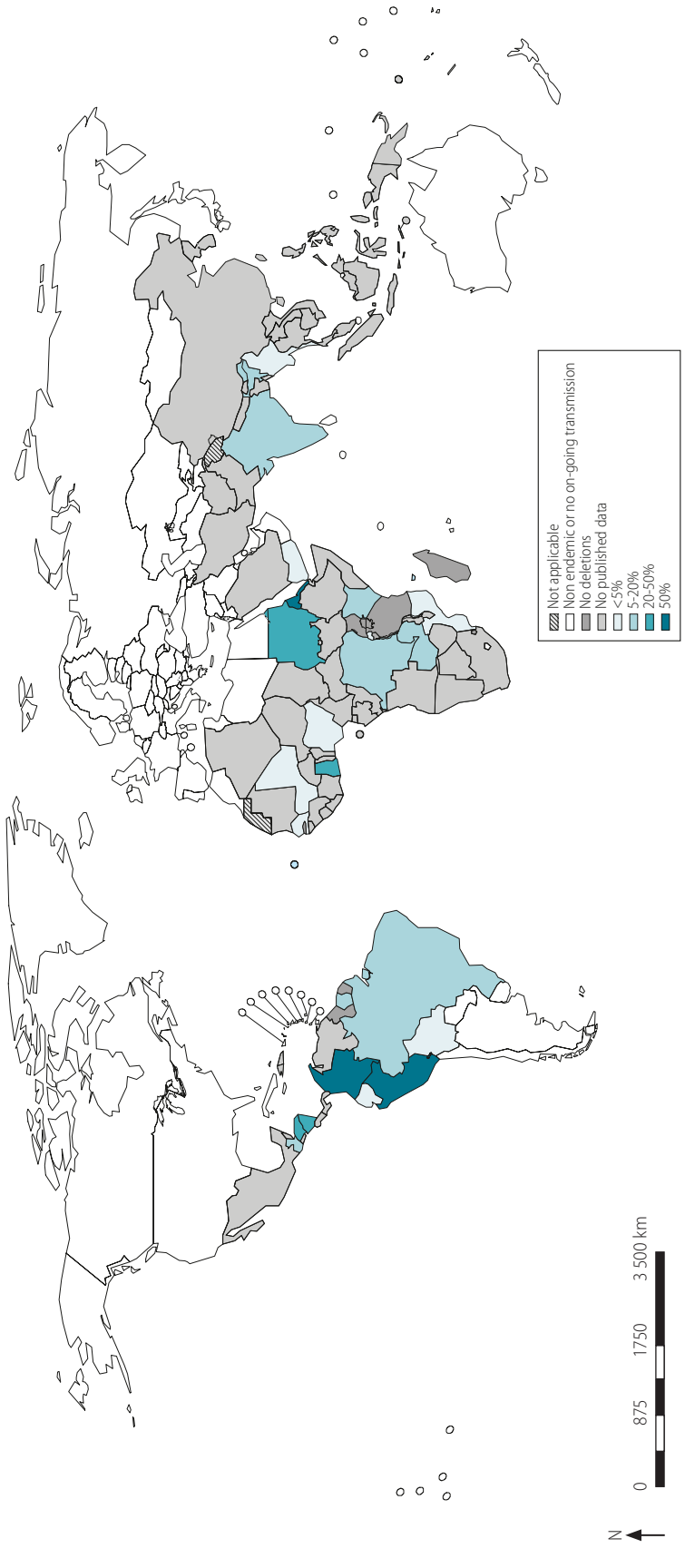
The study has limitations. Although we included only published articles, we were aware of other abstracts and doctoral theses for which relevant data on methods were not available. For manuscripts included in the review, we contacted authors to obtain information not included in the manuscripts; this was not always possible, however, and we therefore occasionally made assumptions about the methods. Survey design and sample populations varied greatly across the included studies. Most studies were not purposely designed for investigating the prevalence of gene deletions and relied on convenience sampling or on secondary analyses of existing specimens. These shortcomings limit our ability to draw conclusions that can inform the use of rapid diagnostic testing, but rather provides guidance for future surveys.

Reconciling the different populations and sample sizes across studies is challenging. First, studies of asymptomatic and symptomatic patients require different interpretations and are difficult to integrate. Samples from asymptomatic patients may have lower parasite densities, resulting in less DNA target for amplification and potentially greater risk of falsely reporting *pfhrp2* deletions. This risk is especially high when the investigation does not include amplification of other single-copy genes or does not quantify parasite DNA. Furthermore, little is known about the effect of *pfhrp2/3* gene deletions on the virulence of malaria infection. If *pfhrp2/3* deletions are associated with less virulent infections, there could be a difference in prevalence between symptomatic and asymptomatic infections. We found numerous studies with low sample sizes which may not be representative of the true prevalence of deletions in a population or country.

Second, different selection criteria for *pfhrp2/3* genotyping (all malaria suspects or only those with discordant HRP2-based rapid diagnostic test and microscopy and/or PCR results) result in the use of different numerators and denominators for prevalence estimation across studies. Analysis limited to deletions found among discordant samples leads to a higher prevalence of gene deletions being reported. To improve consistency in calculating the prevalence of *pfhrp2*-negative mutants, we used the total number of samples with confirmed *pfhrp2* gene deletions by species-specific PCR as the numerator and the total cohort number of *P. falciparum*-positive samples by microscopy and/or PCR as the denominator. The WHO-recommended approach of testing only a subset of high-risk (discordant) samples is a more economical way of monitoring the prevalence of gene deletion and targets clinically significant deletions that cause negative test results. WHO recommends using non-exclusive HRP2-based rapid diagnostic tests when the prevalence of *pfhrp2/3* gene deletions causing false-negative test results is greater than 5%.[22] Most studies included in this review did not allow us to determine if the WHO threshold had been exceeded. It is well acknowledged that the WHO approach may underestimate the prevalence of *pfhrp2* deletions in the parasite population. Samples that are *pfhrp2*-negative and *pfhrp3*-positive are not likely to be flagged as high risk or discordant due to the cross-reactivity between HRP2 and HRP3 proteins on many rapid diagnostic test brands. However, this concern does not pose an immediate threat to patients.[54] Ideally, all *P. falciparum*-positive samples should be used as the denominator, determined either by microscopy or a good quality rapid diagnostic test for detecting *P. falciparum*-specific lactate dehydrogenase.

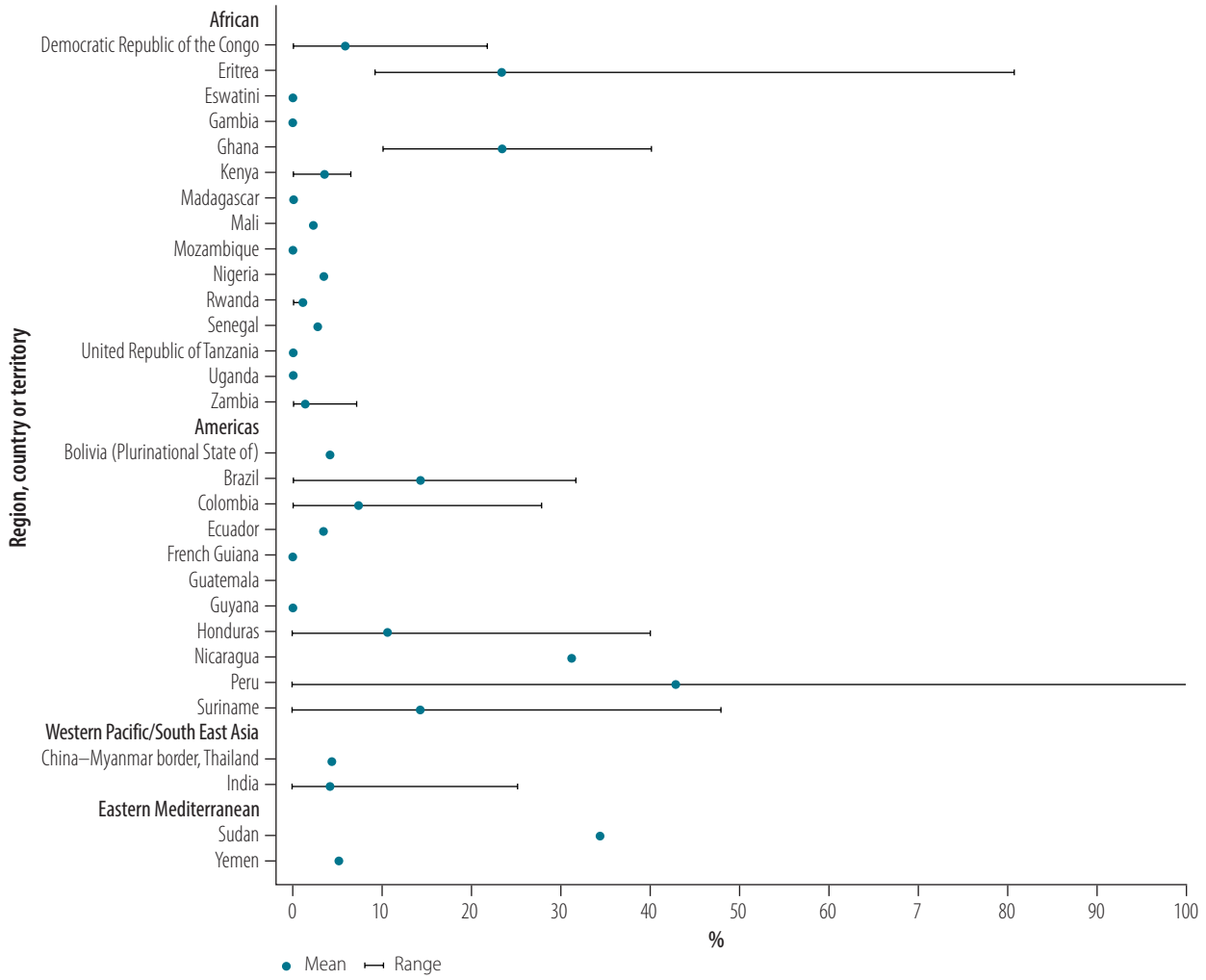
The study bias scores show the potential bias of the prevalence estimates from the true prevalence of *pfhrp2/3* gene deletions in the population, but not necessarily the bias of deletions causing false-negative results (which is more important for determining the effect on malaria case management). Ultimately,

Fig. 2. **Geographical distribution of highest prevalence estimates for *Plasmodium falciparum pfhrp2* gene deletions by study among patients tested at the country level**



Notes: We calculated the prevalence of *pfhrp2* gene deletions using all *Plasmodium falciparum*-positive samples as the denominators by country. We used the highest prevalence estimates for each study.

Fig. 3. **Weighted average prevalence estimates for *Plasmodium falciparum pfhrp2* gene deletions among patients tested by country**



Notes: the prevalence of *phfrp2* gene deletions was calculated using the number of *Plasmodium falciparum*-positive samples as the denominator. In countries with multiple studies prevalence estimates were weighted based on the number of patients tested. Countries with only one study have no bar shown. Cases studies and a study from Zambia that tested for *pfhrp2*-leader sequences are not included.

high-throughput screening options could become more readily available and more commonly used. If so, the true prevalence of *pfhrp2* gene deletions could be determined by molecular testing of all people with suspected malaria regardless of rapid diagnostic test or microscopy results, and those samples confirmed to have *pfhrp2* deletions used as the numerator.

Third, recent modelling suggests that the likelihood of finding *pfhrp2/3* deletions can vary during the malaria transmission season due to changes in the transmission intensity and multiplicity of infection, whereby a person can be infected with multiple *P. falciparum* strains.[55] Co-infection with *pfhrp2/3*-negative- and wild-type parasites can prevent detection of gene deletions using current laboratory techniques, leading to an underestimation of the prevalence of *pfhrp2/3*-negative mutants. Time of year and relation to the transmission season is rarely described in published reports. A publicly available database using prediction models could be useful to help determine the optimal time in the transmission season to conduct a gene deletion survey.[56]

Due to the small number of studies, differing populations and often small samples sizes of the reviewed studies, it is difficult to draw robust conclusions on the prevalence of *pfhrp2/3* gene deletions in specific areas or to perform meta-analysis from these data. The implementation of more large-scale, robust surveys would enable a better understanding of if, and at what rate, these mutations are increasing in a given area, and would allow for meta-analysis.

Identifying the prevalence of *pfhrp2/3* deletion mutations requires synthesis of several lines of evidence and study procedures that include proper performance of rapid diagnostic tests and careful genotyping methods. While most studies in this review followed some components of published criteria to classify a sample as *pfhrp2*-deleted,[21] only 3 (5%) of the studies followed the seven recommended criteria proposed in this review. One specific challenge for molecular analyses of *pfhrp2/3* is using the absence of amplified products as the indicator of interest, rather than the presence of amplified products. Rigorous methods and appropriate controls must be used to ensure the presence of

non-degraded, amplifiable parasite DNA and the lack of amplicon contamination. Improving the accuracy of survey outcomes requires novel molecular-based technology and methods that could detect *pfhrp2/3* gene deletions more reliably and efficiently and detect *pfhrp2/3* deletions in samples with mixed infections (such as quantitative-PCR and whole genome sequencing). Not all malaria-endemic countries have the capacity to conduct molecular analysis to a high standard, and establishing such capacity is challenging and costly. In addition, the sensitivity and specificity of PCR assays can be affected by the protocol used, potentially resulting in variations in the results across laboratories following different procedures. For example, lowering the elongation temperature on *pfhrp2* assays improved the limit of detection of many previously published assays.[57] WHO has established a network of laboratories capable of conducting this analysis to ensure that samples from prevalence surveys can be performed quickly and procedures harmonized across laboratories.[22]

Just over half of the studies amplified both exon 1 and 2 of the *pfhrp2* gene, while the rest amplified only exon 2. While the chromosomal break points could theoretically be anywhere within the *pfhrp2* gene, it appeared that most samples from Eritrea and Peru have observed deletions in both exon 1 and 2 (Qin Cheng, Australian Defence Force Malaria and Infectious Disease Institute, personal communication, 2019). Therefore, whether analysis of exon 2 alone is sufficient to identify most parasites with *pfhrp2* gene deletions requires further analysis of gene deletions from other parasite populations. While not included in the recommendations for *pfhrp2/3* molecular analysis,[21] analysis of flanking genes can provide additional information on genetic mutations.

Despite the diversity of study approaches, there appear to be areas of high prevalence of *pfhrp2/3* mutant parasites where diagnostic testing based on HRP2 alone would be inadequate. Thus, the need for alternative rapid diagnostic tests is of urgent concern in the Amazon basin and Eritrea, where the prevalence of tests producing false-negative results among symptomatic patients has forced changes in the diagnostic strategy.[58] Malaria control programmes should remain vigilant for evidence suggesting the presence of *pfhrp2/3* gene deletions. Evidence of false-negative rapid diagnostic tests or confirmed *pfhrp2/3*-negative mutants in neighbouring countries should trigger careful investigation and surveillance. To improve the quality and relevance of surveys for clinical case management, WHO now provides general guidance on when to prioritize surveys for *pfhrp2/3* deletions.[22] WHO has also developed protocols for guiding survey design, data collection and laboratory methods to determine the prevalence of clinically-relevant *pfhrp2/3* deletions causing false-negative rapid diagnostic tests.[59] The guidelines aim to ensure that future investigations are implemented to high and comparable standards. Additionally, an up-to-date repository of *pfhrp2/3* deletion studies is maintained on the WHO malaria threat map.[60]

The specific factors that drive the evolution and spread of *pfhrp2/3* mutations are not clear, although mathematical models suggest that selective pressure by HRP2-detecting rapid diagnostic tests over the past decade is likely to have played an important role.[5] Low malaria transmission and high frequency of people correctly treated on the basis of diagnosis with HRP2-detecting tests have also been identified as key drivers of the selection of *pfhrp2/3*-negative mutants.[61] Nevertheless, the existence and rising prevalence of *pfhrp2* deletions in Peru,[11,12,33] where HRP2-only methods have never been widely used, along with the local prevalence of *pfhrp3* mutations, confirms that selective treatment based on test results is not the only factor driving the evolution of these parasites.

Due to the global reliance on rapid diagnostic tests for malaria diagnosis, a coordinated, multifaceted response to P. falciparum with *pfhrp2/3* gene deletions is required. This response should include representative studies of the prevalence and distribution of *pfhrp2/3* deletions, more efficient and affordable methods for screening and confirming these deletions, and efforts to standardize and ensure high-quality reporting. Follow-up surveys in areas with documented *pfhrp2/3* deletions will provide insight into the speed at which the mutant parasites are evolving in response to diagnostic pressure and other drivers. Research for the development and commercialization of rapid diagnostic tests based on new or improved non-HRP2 targets is an essential parallel area of work. ■

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Competing interests: None declared.

ملخص

انتشار المتصورة المنجلية (الملاريا الخبيثة) التي تفتقر للبروتينات الغنية بالهيستدين 2 و3: مراجعة منهجية

الغرض حساب تقديرات الانتشار، وتقييم جودة الدراسات التي تشير إلى المتصورة المنجلية (الملاريا الخبيثة) التي تفتقر إلى البروتينات الغنية بالهيستدين 2 و3، وذلك لوضع خطة الاستجابة الدولية.

الطريقة قمنا بالبحث في خمس قواعد للبيانات على الإنترنت، دون التقيد بلغة، عن المقالات التي تحتوي على بيانات أصلية عن المرضى المصابين بالمتصورة المنجلية، مع حذف جينات *pfhrp2* و/أو *pfhrp3* (*pfhrp2/3*). قمنا بحساب تقديرات الانتشار لحالات حذف *pfhrp2/3*، وقمنا بتصنيف البيانات حسب البلد. كان القاسم المشترك هو جميع العينات الإيجابية للمتصورة المنجلية، التي كانت نتيجتها إيجابية بالفحص المجهري، وتأكد أنها إيجابية بواسطة اختبار تفاعل سلسلة بوليميريز النوعي (PCR). إذا لم يتم إجراء الفحص المجهري، فإننا استخدمنا عدد العينات بناءً على طريقة تشخيص مختلفة أو اختبار PCR بمفرده. قمنا بتقييم دراسات لمخاطر التحيز، وجودة طرق المختبر باستخدام نظام تقييم قياسي.

النتائج تضمنت المراجعة إجمالي 38 مقالاً عن 55 دراسة من 32 دولة، ومنطقة واحدة حول العالم. اكتشفنا وجود عدم تجانس ملموس في السكان الذين خضعوا للدراسة، والطرق المستخدمة، والانتشار التقديري لطفيليات المتصورة المنجلية مع حالات حذف *pfhrp2/3*. تراوح الانتشار المترتب لحالات حذف *pfhrp2* من

0% إلى 100%، بما في ذلك المناطق البؤرية في أمريكا الجنوبية وأفريقيا. ثلاث دراسات فقط (5%) استوفت كل المعايير السبعة لجودة الدراسة.
الاستنتاج أدى النقص في الاستقصاءات التمثيلية، أو الاتساق في تصميم الدراسة، إلى إضعاف تقييمات مخاطر النتائج السلبية الكاذبة في تشخيص الملاريا بسبب حالات حذف *pfhrp2/3*. هناك حاجة للتصنيف الدقيق والمراقبة المدعمة لانتشار حالات حذف *pfhrp2/3*، إلى جانب الأساليب المتناغمة التي تسهل المقارنات بين الدراسات.

摘要

缺失富组氨酸蛋白 2 和 3 的恶性疟原虫感染率：系统评审

目的 旨在计算感染率估值并评估报告缺失富组氨酸蛋白 2 和 3 的恶性疟原虫的研究质量，以制定一项国际性应对计划。
方法 我们在没有语言限制的情况下搜索了五个在线数据库，以获取报告缺失 *pfhrp2* 和 / 或 *pfhrp3* 基因 (*pfhrp2/3*) 的恶性疟原虫感染患者原始数据的文章。我们计算了缺失 *pfhrp2/3* 的感染率估值并按国家映射数据。分母是所有经显微镜检测均为阳性并通过种特异性聚合酶链反应试验 (PCR) 确认为阳性的恶性疟原虫阳性样本。如果进行显微镜检测，我们采用基于不同诊断方法或单独 PCR 的样本数量。我们使用标准化评分系统对偏倚风险以及实验室方法的质量研究进行了评分。
结果 评审中共纳入 38 篇文章，这些文章报告了全球 32 个国家和 1 个地区中的 55 项研究。在研究的人群、使用的方法以及估计的缺失 *pfhrp2/3* 的恶性疟原虫感染率中，我们发现了相当大的异质性。推导的 *pfhrp2* 缺失感染率介于 0% 至 100% 之间，包括南美洲和非洲的重点地区。就研究质量而言，只有三项研究 (5%) 符合所有七个标准。
结论 缺乏代表性调查或研究设计缺乏一致性妨碍了由于 *pfhrp2/3* 缺失导致疟疾诊断中假阴性结果风险的评估。需要准确映射和加强对 *pfhrp2/3* 缺失感染率的监测，以及促进各项研究比较的统一方法。

Résumé

Prévalence de *Plasmodium falciparum* sans protéines 2 et 3 riches en histidine: revue systématique

Objectif Estimer la prévalence et évaluer la qualité des études consacrées à *Plasmodium falciparum* sans protéines 2 et 3 riches en histidine afin d'établir un plan d'intervention international.
Méthodes Nous avons parcouru cinq bases de données en ligne sans restriction de langue pour trouver des articles contenant des informations d'origine relatives à des patients atteints de *Plasmodium falciparum* dépourvu des gènes *pfhrp2* et/ou *pfhrp3* (*pfhrp2/3*). Nous avons calculé la prévalence des délétions des gènes *pfhrp2/3* et cartographié les données par pays. Le dénominateur était représenté par les échantillons positifs à *P. falciparum*, testés positifs au microscope et confirmés par un test de réaction en chaîne par polymérase (PCR) propre à l'espèce. Si aucun examen n'avait été effectué au microscope, nous avons utilisé le nombre d'échantillons recourant à une méthode de diagnostic différente, ou uniquement à la PCR. Nous avons noté les études selon le risque de biais et la qualité des techniques d'analyse en laboratoire, à l'aide d'un système de notation standardisé.
Résultats Au total, 38 articles mentionnant 55 études réalisées dans 32 pays et un territoire dans le monde ont été pris en compte dans cette revue. Nous avons observé une grande hétérogénéité dans les populations étudiées, les méthodes employées et la prévalence estimée des parasites *P. falciparum* assortis d'une délétion des gènes *pfhrp2/3*. La prévalence dérivée des délétions de *pfhrp2* est comprise entre 0% et 100%, avec des zones de convergence en Amérique du Sud et en Afrique. Seules trois études (5%) remplissaient l'ensemble des sept critères de qualité.
Conclusion L'absence d'enquêtes représentatives ou d'uniformité dans la conception des études empêche d'évaluer correctement le risque de faux négatifs dans le diagnostic de la malaria en raison des délétions de *pfhrp2/3*. Une cartographie détaillée ainsi qu'une surveillance renforcée de la prévalence des délétions de *pfhrp2/3* est nécessaire, tout comme une harmonisation des méthodes afin de faciliter la comparaison entre les différentes études.

Резюме

Распространенность *Plasmodium falciparum* с отсутствием богатых гистидином белков 2 и 3: систематический обзор

Цель Вычислить распространенность и оценить качество исследований, посвященных распространенности *Plasmodium falciparum* с отсутствием богатых гистидином белков 2 и 3, с целью получения информации для разработки международного плана реагирования.
Методы Авторы выполнили поиск статей, содержащих исходные данные по пациентам, зараженным *Plasmodium falciparum* с делецией генов *pfhrp2* и/или *pfhrp3* (*pfhrp2/3*), в пяти базах данных в Интернете на разных языках. Авторы выполнили оценку распространенности делеций *pfhrp2/3* и составили карту данных для разных стран. В качестве знаменателя использовалось общее количество проб на *P. falciparum* с положительным результатом, полученным при микроскопическом исследовании и подтвержденным в ходе видоспецифичного тестирования методом полимеразной цепной реакции (ПЦР). Если микроскопическое исследование не проводилось, авторы использовали данные о количестве проб, полученные на основании другого диагностического метода или только на основании ПЦР. Исследования оценивались по уровню риска необъективности и качеству лабораторных методов с применением стандартной системы оценок.

Результаты В общей сложности в обзор были включены 38 статей, содержащих сведения о 55 исследованиях, проведенных в 32 странах мира и на одной территории. Авторы обнаружили значительную неоднородность в исследованных популяциях, использованных методах и оценке распространенности паразитов *P. falciparum* с делециями *pfhrp2/3*. Производное значение распространенности делеций *pfhrp2* находилось в диапазоне от 0 до 100%, включая очаговые области в Южной Америке и Африке. Только три исследования (5%) соответствовали всем семи критериям качества для исследований.

Вывод Отсутствие репрезентативных исследований или недостаточное единообразие планов исследований отрицательно влияют на оценку риска ложноотрицательных результатов при диагностировании малярии по причине делеций *pfhrp2/3*. Необходимы точное отображение данных и усиленный мониторинг распространенности делеций *pfhrp2/3*, а также разработка гармонизированных методов, которые упростят сравнение разных исследований между собой.

Resumen

Prevalencia del *Plasmodium falciparum* que carece de las proteínas 2 y 3 ricas en histidina: un análisis sistemático

Objetivo Calcular las estimaciones de la prevalencia y evaluar la calidad de los estudios que informan de la existencia del *Plasmodium falciparum* que carece de las proteínas 2 y 3 ricas en histidina, para elaborar un plan de respuesta internacional.

Métodos Se revisaron cinco bases de datos en línea, sin restricción de idioma, para encontrar artículos que informaran sobre los datos originales de los pacientes infectados con *Plasmodium falciparum* con deleciones de los genes *pfhrp2* y/o *pfhrp3* (*pfhrp2/3*). Se calcularon las estimaciones de prevalencia de las deleciones de *pfhrp2/3* y se clasificaron los datos por país. El denominador eran todas las muestras positivas por *P. falciparum* que daban positivo en las pruebas de microscopía y confirmadas como positivas en las pruebas de reacción en cadena de la polimerasa (PCR, por sus siglas en inglés) específicas de la especie. Si no se realizaba la microscopía, se empleaba el número de muestras en base a un método de diagnóstico diferente o a la PCR únicamente. Los estudios se calificaron en función del riesgo de sesgo y de la calidad de los métodos de laboratorio por medio de un sistema de puntuación estandarizado.

Resultados El análisis incluyó un total de 38 artículos en los que se informaba de 55 estudios de 32 países y un territorio a nivel mundial. Se observó una heterogeneidad considerable en las poblaciones estudiadas, los métodos aplicados y la prevalencia estimada de los parásitos *P. falciparum* con deleciones de los genes *pfhrp2/3*. La prevalencia que se estimó de las deleciones del gen *pfhrp2* osciló entre el 0 % y el 100 %, incluidas las áreas focales de América del Sur y África. Tan solo tres estudios (5 %) cumplieron los siete criterios de calidad del estudio.

Conclusión La falta de encuestas fiables o de consistencia en el diseño de los estudios dificulta las evaluaciones del riesgo de resultados falsos negativos en el diagnóstico de la malaria debido a las deleciones de los genes *pfhrp2/3*. Se necesita un mapeo preciso y un seguimiento reforzado de la prevalencia de las deleciones de los genes *pfhrp2/3*, junto con métodos estandarizados que faciliten las comparaciones entre los estudios.

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Table 2. **Studies reporting *pfhrp2 and pfhrp3* gene deletions and derived prevalence estimates by region in the systematic review of *Plasmodium falciparum pfhrp2/3* gene deletions**

Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Africa													
Koita et al., 2012[41]	Mali	1996	Sirakoro, Bancoumana, Doneguebougou, Bamako	Cross-sectional	Mixed	Discordant	480	10[d] (2)	NA	NA	NA	3	3
Ramutton et al., 2012[35]	Democratic Republic of the Congo	2005–2010	Kinshasa	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	6[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	Gambia	2005–2010	Banjul	Health facility (Antimalarial drug trial)	Unrepresentative	Subsample	2[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	Kenya	2005–2010	Kilifi	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	12[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	Mozambique	2005–2010	Beira	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	19[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	Rwanda	2005–2010	Kigali, Nyanza	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	15[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	United Republic of Tanzania	2005–2010	Teule, Korogwe	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	18[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Ramutton et al., 2012[35]	Uganda	2005–2010	Mbarare	Health facility (antimalarial drug trial)	Unrepresentative	Subsample	5[e]	0 (0)	0 (0)	0 (0)	NA	5	2
Wurtz et al., 2013[43]	Senegal	2009–2012	Dakar	Health facility (antimalarial drug trial)	Symptomatic	All	125	3 (2)	16 (13)	NA	NA	5	2
Laban et al., 2015[45]	Zambia	2008–2012	Choma	Cross-sectional	Mixed	All	61	NA	NA	12[f] (20)	NA	4	1
Amoah et al., 2016[16]	Ghana	2015	Abura Dunkwa, Obom	Malaria screening programme	Mixed	All	288	76 (26)	85 (30)	37 (13)	22–40	6	1

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Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Parr et al., 2017[15]	Democratic Republic of the Congo	2013–2014	Kinshasa, Kwango, Kwilu, Mai-Ndombe, Kongo Central, Equateur, Mongala, Nord-Ubangi, Sud-Ubangi, Tshuapa, Kasai, Kasai-Central, Kasai-Oriental, Lomami, Sankuru, Haut-Katanga, Haut-Lomami, Lualaba, Tanganyka, Maniema, Nord-Kivu, Bas-Uele, Haut-Uele, Ituri, Tshopo, Sud-Kivu	Cross-sectional	Mixed	Discordant	2752[g]	149[h] (5)[i]	NA	5 (< 1)	0–22	5	3
Beshir et al., 2017[9]	Kenya	2014	Mbita	Cross-sectional in schools (mosquito behaviour study)	Asymptomatic	All	131	8 (6)	1 (1)	0 (0)	NA	6	2
Beshir et al., 2017[9]	Kenya	2007–2008	Kilifi	Health facility	Symptomatic	All	49	1 (2)	1 (2)	0 (0)	NA	4	2
Gupta et al., 2017[17]	Mozambique	2010–2016	Manhiça, Magude	Cross-sectional	Mixed	Discordant	1162	1[j] (< 1)	0 (0)	0 (0)	NA	6	3
Kozycki et al., 2017[14]	Rwanda	2014–2015	Busogo, Kiribizi, Bukara	Health facility	Symptomatic	Discordant	3291	32[k] (1)	NA	NA	NA	4	4
Menegon et al., 2017[36]	Eritrea	2013–2014	Gash Barka, Debun	Unknown	Unknown	All	144	14 (10)	62 (43)	13 (9)	9–22	4	2
Ranadive et al., 2017[37]	Eswatini	2012–2014	Lubombo	Health facility	Symptomatic	Discordant	162[c]	0[l] (0)	1 (1)	0 (0)	NA	4	2
Berhane et al., 2018[20]	Eritrea	2016	Northern Red Sea, Anseba, Gash Barka, Debub	Health facility	Symptomatic	All	50	31 (62)	41 (82)	31 (62)	42–81	7	2

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Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Nderu et al., 2018[39]	Kenya	2007–2016	Busia, Mbita, Nyando, Tiwi, Msambweni	Drug efficacy trial	Symptomatic	All	400	0 (0)	0 (0)	0 (0)	NA	5	2
Owusu et al., 2018[38]	Ghana	2015	Greater Accra, Eastern region	Cross-sectional study among patients attending antiretroviral therapy clinics	Unrepresentative	Discordant	62	6[m](10)	8 (13)	6 (10)	NA	4	3
Willie et al., 2018[40]	Madagascar	2014–2015	Yurimaguas	Health facility	Symptomatic	All	73	0 (0)	NA	NA	NA	3	2
Funwei et al., 2019[42]	Nigeria	2013–2014	Elata Ibadan	Health facility	Symptomatic	Discordant	340	11n (3)	4 (1)	11 (3)	NA	6	4
Kobayashi et al., 2019[46]	Zambia	2009–2011	Choma	Cross-sectional	Mixed	Discordant	45	3[o] (7)	NA	0 (0)	NA	5	3
Kobayashi et al., 2019[46]	Zambia	2015–2017	Nchelenge	Cross-sectional	Mixed	Discordant	1144	0[p] (0)	NA	0 (0)	NA	6	3
Americas													
Gamboa et al., 2010[11]	Peru	2003–2007	Iquitos area, Loreto, Amazonas, Cajamarca	Health facility	Unknown	All	148	61 (41)	103 (70)	31 (22)	36–100	4	2
Gamboa et al., 2010[11]	Peru	2007	Iquitos	Active case detection survey	Symptomatic	All	9	8 (90)	6 (67)	4 (44)	NA	7	2
Houzé et al., 2011[25]	Brazil	2011	Amazon region	Case study	Symptomatic	Discordant	1	1[q]	1[q]	1[q]	NA	5	4
Maltha et al., 2012[12]	Peru	2010–2011	Iquitos area	Health facility, active case detection	Symptomatic	All	74	19 (26)	34 (44)	19 (26)	NA	6	2
Akinyi et al., 2013[13]	Peru	1998–2001	Loreto, Piura	Unknown	Symptomatic	All	92[c]	19 (21)	NA	NA	0–36	2	2
Akinyi et al., 2013[13]	Peru	2003–2005	Iquitos	Unknown	Symptomatic	All	96[c]	39 (41)	NA	NA	NA	2	2

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Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Trouvay et al., 2013[29]	French Guiana	2009	St Luarent du Maroni, Cayenne, St Georges de l'Oyapack, Saul, Antecume Pata	Unknown	Symptomatic	All	140[c]	0 (0)	4 (3)	0 (0)	NA	2	2
Trouvay et al., 2013[29]	French Guiana	2010–2011	Cayenne Hospital	Health facility survey	Symptomatic	All	81	0 (0)	6 (7)	0 (0)	NA	5	2
Abdallah et al., 2015[32]	Honduras	2008–2009	Puerto Lempira	Health facility (antimalarial drug trial)	Symptomatic	All	68[c]	0 (0)	30 (44)	0 (0)	NA	2	2
Akinyi Okoth et al., 2015[31]	Guyana	2009–2011	Georgetown (Cuyuni-Mazaruni, Potaro-Siparuni)	Health facility survey	Symptomatic	All	97	0 (0)	0 (0)	0 (0)	NA	3	2
Akinyi Okoth et al., 2015[31]	Suriname	2009–2011	Sipaliwini, Brokopondo	Health facility survey, active case detection	Symptomatic	All	78	11 (14)	3 (4)	2 (3)	0–48	3	2
Baldeviano et al., 2015[33]	Peru	2010–2012	Tumbes	Health facility during malaria outbreak	Symptomatic	All	54	54 (100)	NA	NA	NA	2	2
Murillo Solano et al., 2015[26]	Colombia	2008–2009	Cordoba, Narino, Valle del Cauca,	Unknown	Symptomatic	All	75	4 (5)	40 (53)	4 (5)	0–33	6	2
Murillo Solano et al., 2015[26]	Colombia	1999–2007	Amazonas, Guaviare, Meta	Epidemiological studies	Symptomatic	All	25	14 (56)	12 (48)	9 (36)	0–67	4	2
Sáenz et al., 2015[28]	Ecuador	2012–2013	Esmereldas	Malaria outbreak surveillance	Symptomatic	All	32	1 (3)	1 (3)	1 (3)	NA	4	2
Dorado et al., 2016[27]	Colombia	2003–2010	Antiquia, Amazonas, Guaviare, Narino, Choco, Cauca, Valle	Unknown	Symptomatic	All	253	15 (6)	106 (42)	15 (6)	0–54	4	2
Dorado et al., 2016[27]	Colombia	2011–2012	Antiquia, Amazonas, Guaviare, Narino, Choco, Cauca, Valle	Health facility survey	Symptomatic	All	112	0 (0)	51 (42)	0 (0)	NA	6	2

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Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Okoth et al., 2016[34]	Peru	2013	Cusco	Outbreak surveillance	Symptomatic	All	4	4 (100)	4 (100)	4 (100)	NA	4	2
Rachid Viana et al., 2017[24]	Bolivia (Plurinational State of)	2010–2012	Beni department	Health facility survey	Symptomatic	All	25[c]	1 (4)	17 (68)	0 (0)	NA	3	2
Rachid Viana et al., 2017[24]	Brazil	2010–2012	Acre, Para, Rondonia	Health facility survey	Symptomatic	All	198	27 (14)	71 (36)	43 (23)	0–32	4	2
Fontecha et al., 2018[30]	Guatemala	2015	Escuintla	Malaria surveillance survey	Symptomatic	All	21	3 (14)	19 (91)	3 (14)	NA	4	2
Fontecha et al., 2018[30]	Honduras	2011–2017	Gracias a Dios, Colon, Antantida, Cortes, Islas de la Bahia	Health facility survey for drug resistance	Symptomatic	All	52	13 (25)	50 (96)	13 (25)	0–40	4	2
Fontecha et al., 2018[30]	Nicaragua	2015	North Atlantic Autonomous Region	Malaria surveillance survey	Symptomatic	All	55	17 (31)	48 (87)	11 (20)	NA	4	2
South-East Asia													
Kumar et al., 2013[49]	India	2010	Chhattisgarh	Unknown	Symptomatic	All	48[c]	2 (4)	2 (4)	2 (4)	NA	6	2
Li et al., 2015[48]	China–Myanmar border, Thailand[r]	2011–2012	China, Myanmar border and Tak province, Thailand	Mass blood survey, unknown	Unknown	All	97	4 (4)	3[s] (3)	3 (3)	NA	5	2
Bharti et al., 2016[18]	India	2014	Odisha, Chhattisgarh, Jharkhand, Madhya Pradesh, Maharashtra, Rajasthan, Gujarat, Tripura	Health facility	Symptomatic	Discordant	1521	36[t] (2)	27 (2)	25 (2)	0–25	6	4
Nima et al., 2017[47]	Bangladesh	2013	Sylhet	Case study	Symptomatic	Discordant	1	1[q]	1[q]	1[q]	NA	5	4
Pati et al., 2018[50]	India	2013–2016	Odisha	Cross-sectional	Symptomatic	Discordant	384	38[u] (10)	24 (6)	17 (4)	8–14	7	4
Eastern Mediterranean													

(*continues. . .*)

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Region and study	Country or territory	Year of data collection	Study sites	Study design	Sample population[a]	Samples tested[b]	Total no. of *P. falciparum* positive patients[c]	No. (%) of samples with gene deletions			Range of *pfhrp2* deletion prevalence across study sites	Quality score	Study bias score
								Pfhrp2	*Pfhrp3*	*Pfhrp2* and *3*			
Atroosh et al., 2015[19]	Yemen	2014	Hodeidah, Al-Mahwit	Active case detection	Symptomatic	All	189	9 (5)	NA	NA	NA	4	2
Mussa et al., 2019[44]	Sudan	Unrepresentative	Omdurman	Health facility	Symptomatic	All	26	9 (35)	NA	NA	NA	2	2

DNA: deoxyribonucleic acid; HRP2: histidine-rich protein 2; NA: not applicable; PCR: polymerase chain reaction.

[a] Symptomatic: only symptomatic people tested; Mixed: mix of symptomatic and asymptomatic people tested; Asymptomatic: only asymptomatic people tested; Unrepresentative: a subset of people not representative of the population were tested; Unknown: not reported.

[b] All: all samples underwent molecular analysis; Discordant: only discordant samples tested; Subsample: another subset of samples tested.

[c] As microscopy was not performed, we used the number of *P. falciparum* positive cases by PCR as the denominator.

[d] Only 22 samples which were rapid diagnostic test-negative and microscopy-positive samples were analysed for *pfhrp2* deletion.

[e] Samples testing positive by *P. falciparum*-specific lactate dehydrogenase rapid diagnostic test and confirmed by PCR. Only those samples with the lowest level of HRP2 were analysed with molecular methods.

[f] Only the HRP-leader sequence shared by both *pfhrp2* and *pfhrp3* genes was tested, so we could not break down results by *pfhrp2* or *pfhrp3* genes.

[g] Microscopy-positive and -negative samples were analysed. PCR-positive samples are included here.

[h] Only 783 samples which were rapid diagnostic test-negative and PCR-positive were analysed for *pfhrp2* deletion.

[i] Results differ from those presented in the article as we presented unweighted results.

[j] Only 69 samples which were rapid diagnostic test-negative and microscopy-positive were analysed for *pfhrp2* deletion.

[k] Only 138 samples which were rapid diagnostic test-negative and microscopy-positive were analysed for *pfhrp2* deletion.

[l] Only nine samples which were rapid diagnostic test-negative and quantitative PCR-positive with a parasite counts > 100 per µL were analysed for *pfhrp2* deletion.

[m] Only eight samples which were rapid diagnostic test-negative and PCR positive were analysed for *pfhrp2* deletion.

[n] Only 66 samples which were rapid diagnostic test-negative and microscopy- or PCR-positive were analysed for *pfhrp2* deletion.

[o] Only eight samples which were rapid diagnostic test-negative and quantitative PCR-positive with *pfldh* DNA concentration >0.0001 ng per µL *P. falciparum* DNA were analysed for *pfhrp2* deletion.

[p] Only 28 samples which were rapid diagnostic test-negative and microscopy-positive with *pfldh* DNA concentration > 0.0001 ng per µL *P. falciparum* DNA were analysed for *pfhrp2* deletion.

[q] We did not present prevalence for case studies.

[r] Article reported collection of samples from three countries but did not present results separately, so the results have been presented here as one study.

[s] 97 samples were tested for *pfhrp2* deletion, however only the four negative samples were analysed for *pfhrp3* deletion. We present results out of the 97 samples analysed.

[t] Only 50 samples which were rapid diagnostic test-negative and microscopy-positive were analysed for *pfhrp2* deletion.

[u] Only 58 samples which were rapid diagnostic test-negative and microscopy-positive were analysed for *pfhrp2* deletion.

Notes: We calculated the prevalence of gene deletions using all *Plasmodium falciparum*-positive samples as the denominators. All studies used microscopy with PCR confirmation, except where indicated. In some cases, only rapid diagnostic test-negative, microscopy-positive samples or rapid diagnostic test-negative, PCR-positive samples were analysed for *pfhrp2* gene deletion, as indicated in footnotes above. We scored study quality from 1 (lowest) to 7 (highest), as described in Box 1, and study bias from 1 (lowest) to 4 (highest).

The threat of *pfhrp2/3* deletions in the Horn of Africa

Jane Cunningham, Medical Officer

MPAC April 2021

Global **Malaria** Programme

World Health Organization

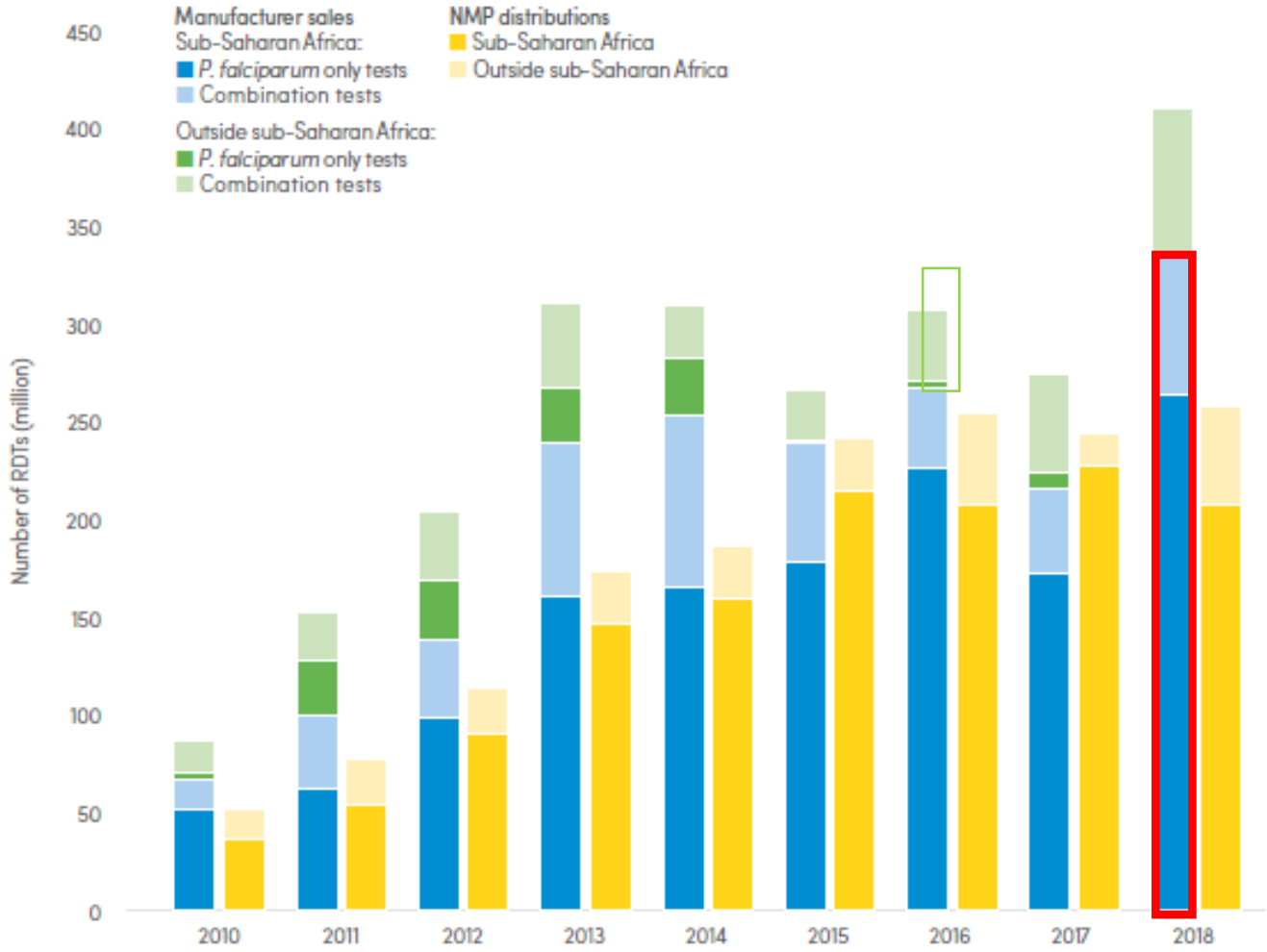
Background

- RDTs target a range of malaria antigens

	HRP2	pLDH	Aldolase
***P.falciparum*-specific**	+	+	
Pan-specific (all species)		+	+
***P.vivax*-specific**		+	

- The majority of RDTs used to detect *P. falciparum* target histidine rich protein-2

Number of RDTs sold by manufacturers and distributed by NMPs for use in testing suspected malaria cases,[a] 2010–2018 *Sources: NMP reports and sales data from manufacturers eligible for WHO's Malaria RDT Product Testing Programme.*



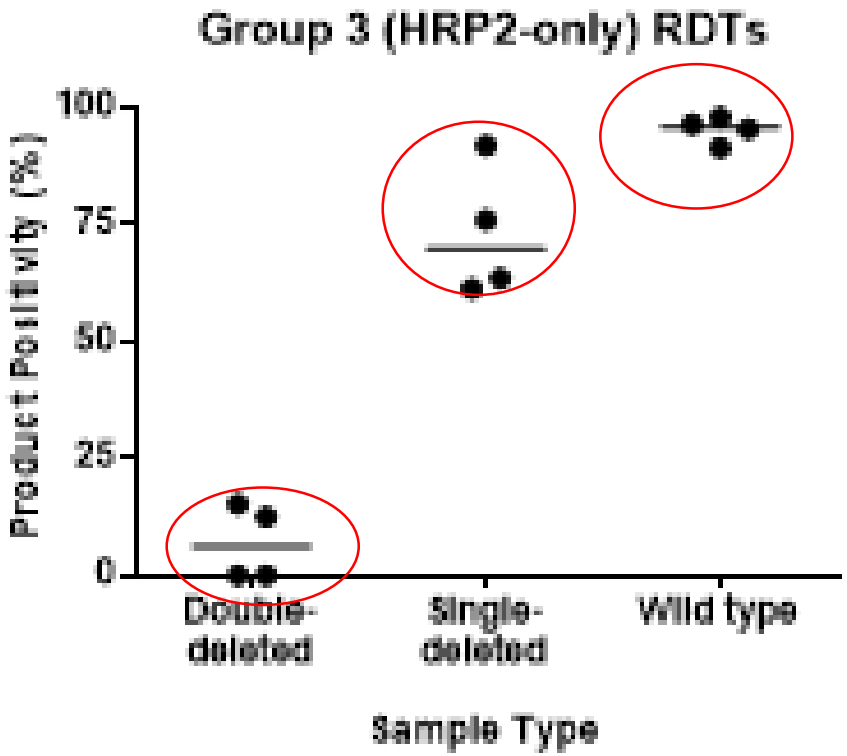
NMP: national malaria programme; *P. falciparum*: *Plasmodium falciparum*; RDT: rapid diagnostic test; WHO: World Health Organization.
[a] NMP distributions do not reflect those RDTs still in storage that have yet to be delivered to health facilities and community health workers.

Background

- HRP2 is found in the cytoplasm and surface of Pf-infected erythrocytes; its produced in abundance but function is not very well understood (number of theories) and Pf parasites can survive without it

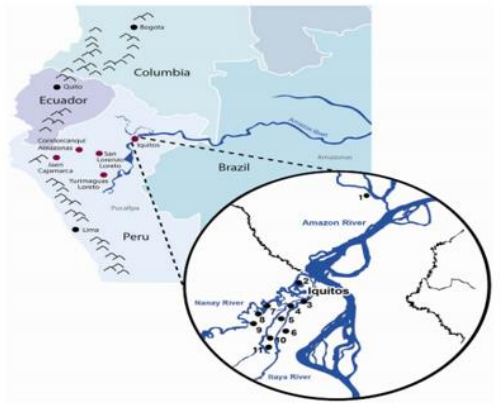
- HRP3 close cousin – share common epitopes

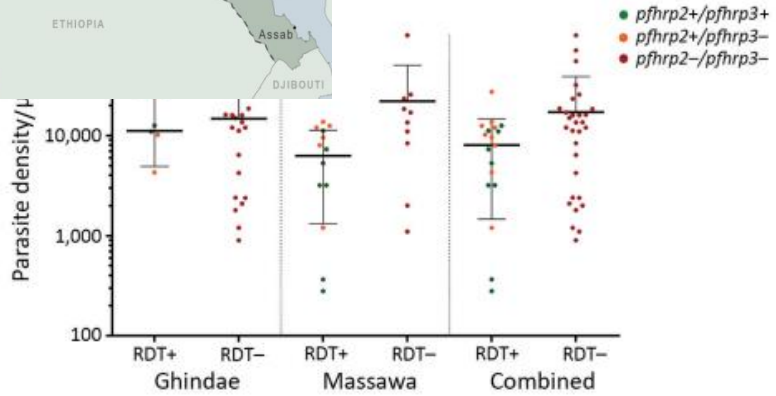
- RDTs that target HRP2 can to some extent also detect HRP3



Source: Malaria RDT Test performance: WHO Product Testing Round 8 (2016-2018)

First reports in Peru in 2010 …Turning point in 2016





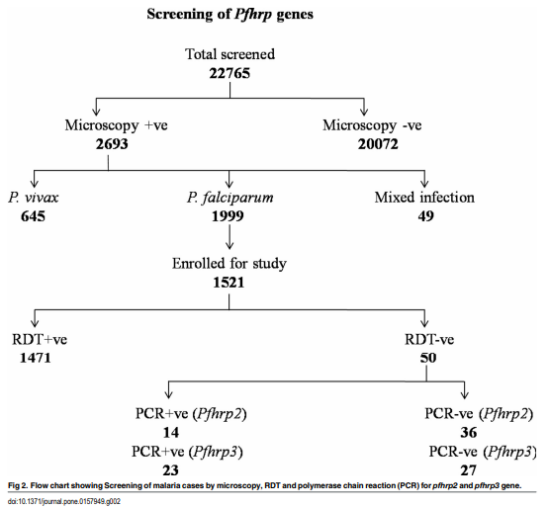


Fig 2. Flow chart showing Screening of malaria cases by microscopy, RDT and polymerase chain reaction (PCR) for *pfhrp2* and *pfhrp3* gene.

doi:10.1371/journal.pone.0157949.g002

- 41% (61/148) of isolates lacked *pfhrp2;*
- 21% lacked both *pfhrp2 and 3*

Very high prevalence of double deletions in Eritrea and overall low but heterogeneous prevalence of deletions in India (eight states)

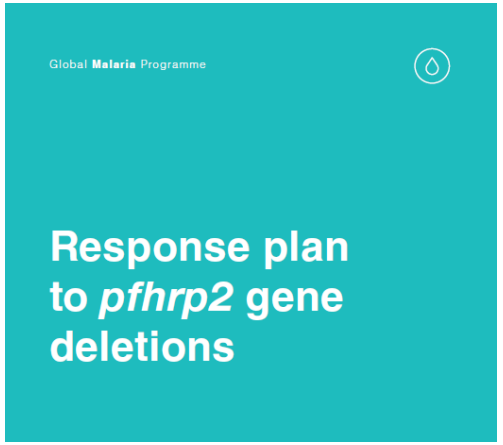
Berhane A, et al. Major Threat to Malaria Control Programs by Plasmodium falciparum Lacking Histidine-Rich Protein 2, Eritrea. Emerg Infect Dis. 2018 Mar;24(3):462-470.

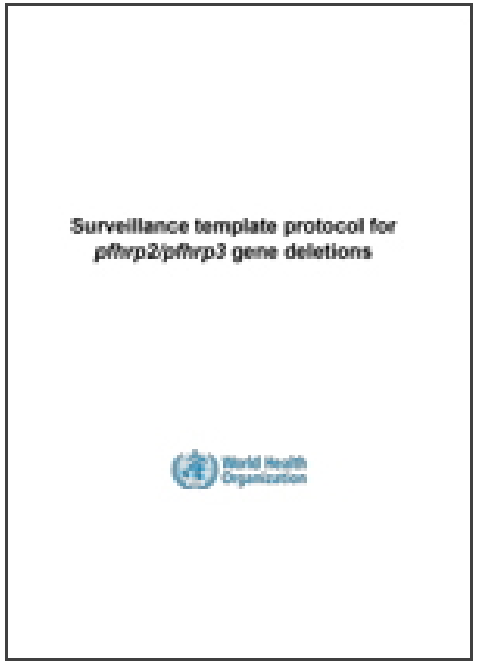
Bharti PK et al (2016) Prevalence of pfhrp2 and/or pfhrp3 Gene Deletion in Plasmodium falciparum Population in Eight Highly Endemic States in India. PLoS ONE 11(8): e0157949.

WHO Response Plan to *pfhrp2/3* deletions

Core response plan to pfhrp2/3 deletions

- ✗ **mapping the distribution and frequency of *pfhrp2/3* deletion mutants with harmonized protocols;**
- ✓ building an international network of laboratories to perform the complex molecular confirmation required for mapping and identify new and/or efficient screening methods ;
- ✓ supporting countries in the selection and procurement of new RDTs when a change of testing is warranted;
- ✓ advising commercial manufacturers of the priorities for new tests and providing the best available market forecasts;

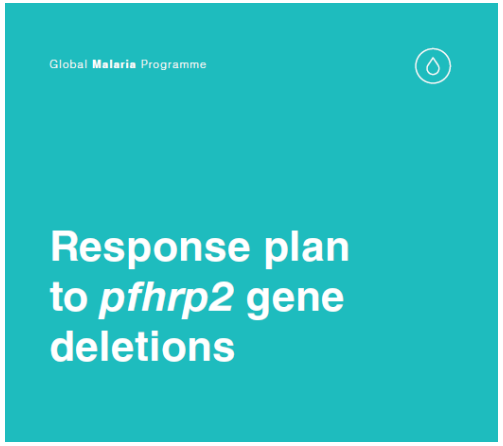


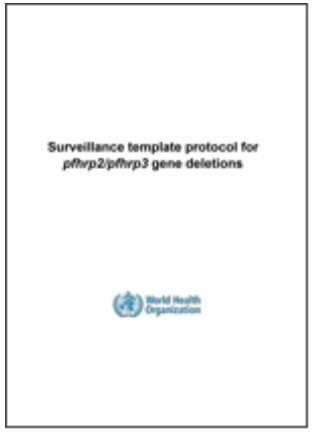


https://apps.who.int/iris/bitstream/handle/10665/325528/WHO-CDS-GMP-2019.02-eng.pdf?sequence=1&isAllowed=y
https://www.who.int/malaria/publications/atoz/hrp2-deletion-protocol/en/

Criteria for change

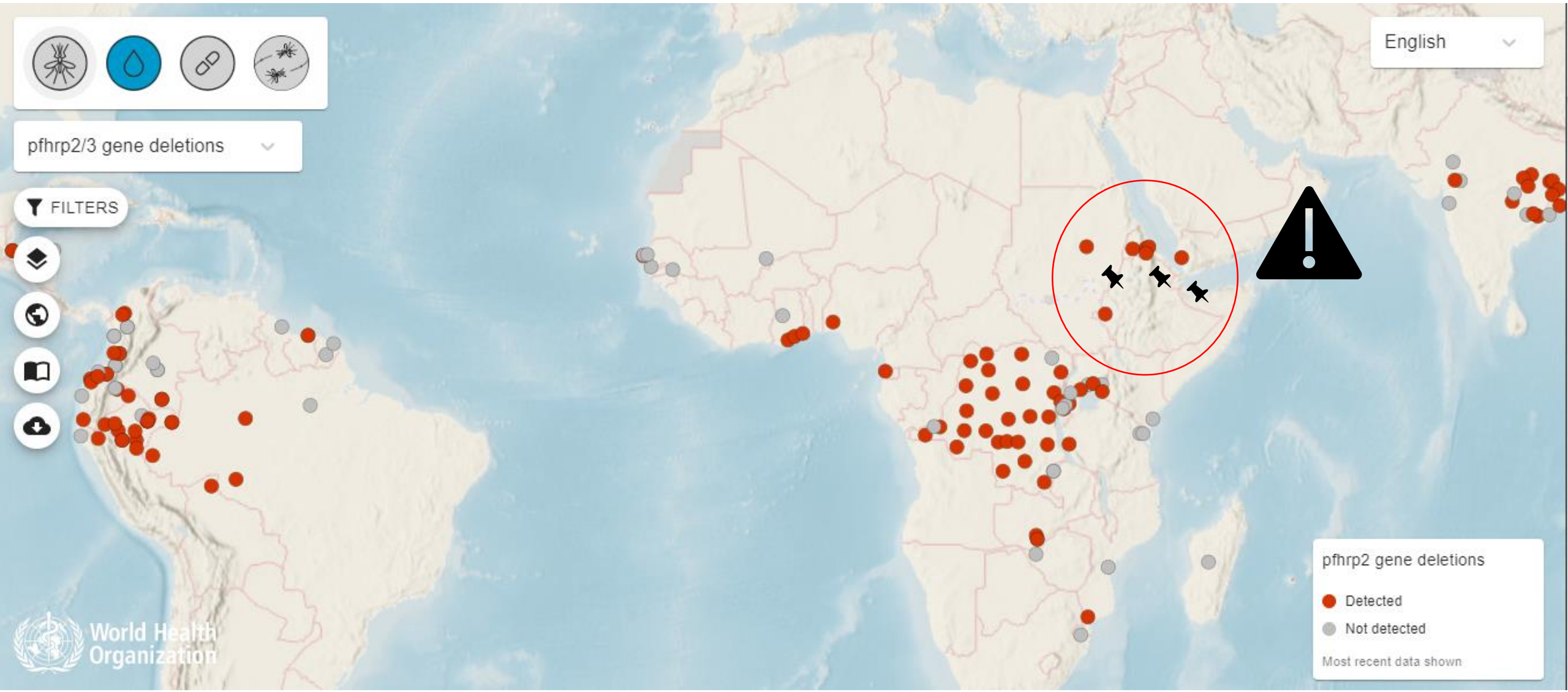
- If a survey confirms the presence of ***pfhrp2/3 deletions causing false negative HRP2-RDTs is greater than 5%*** then the NMCP will need to take a series of actions to immediately optimize case management and plan for the introduction of replacement RDTs. ***Any change should be applied nationwide, although roll-out might be prioritized on the basis of the prevalence of pfhrp2 deletions.***





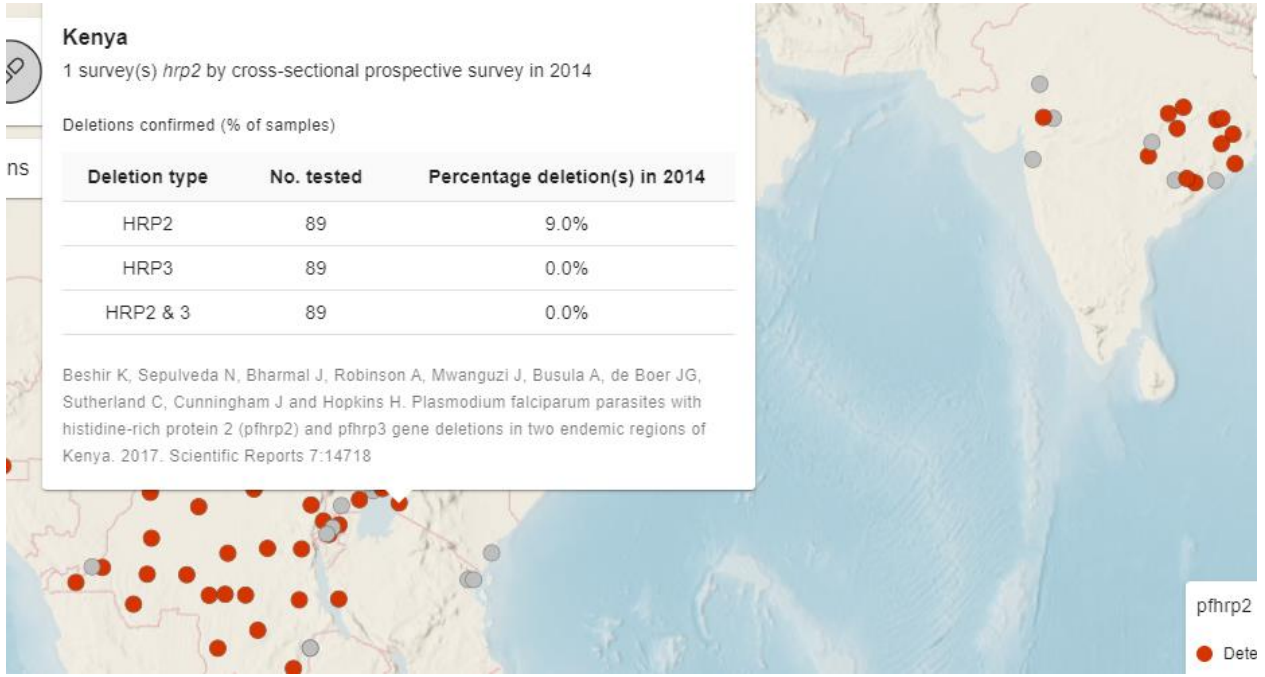
WHO Malaria Threat Maps

https://www.who.int/malaria/maps/threats-about/en/


English
pfhrp2/3 gene deletions
FILTERS
World Health Organization
pfhrp2 gene deletions
Detected
Not detected
Most recent data shown

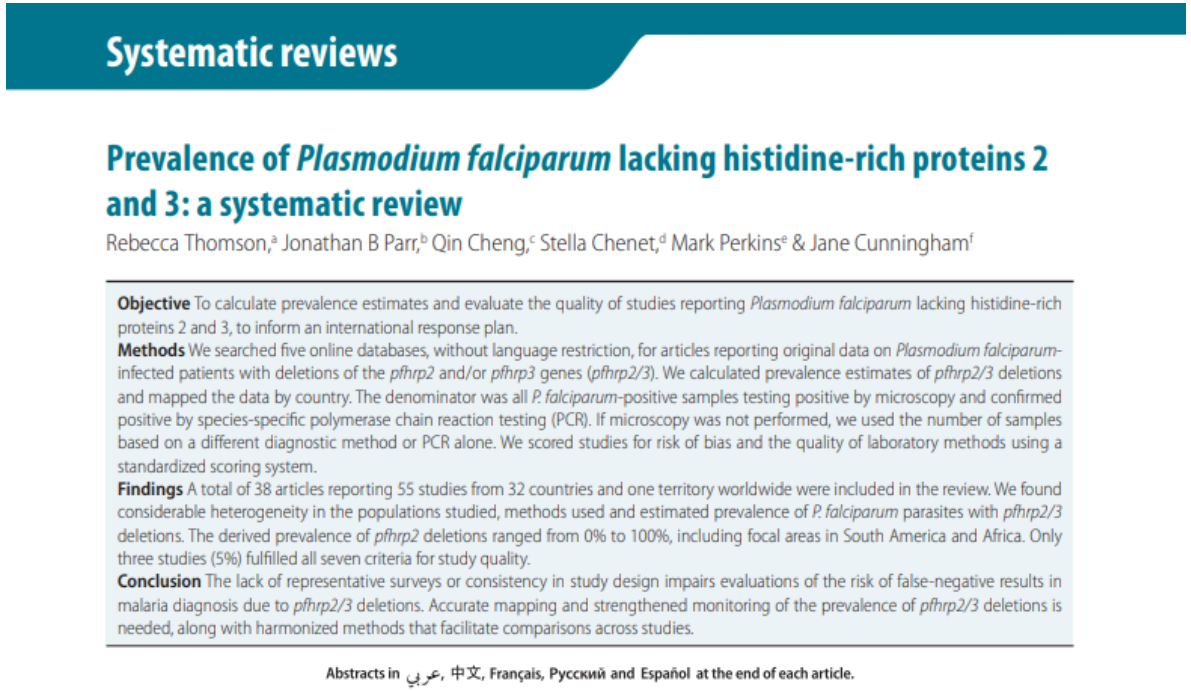
Getting at the true picture ….

- Malaria threat maps chart what is in the published report – typically percentage of pfhrp2 deleted samples amongst those tested and NOT all *P.falciparum* cases
- Populations are different – age, symptoms/no symptoms, selection criteria for genotyping
- RDT result not always known – don't know if the deletion led to a false negative result
- Original source is required to properly interpret the results.



Getting at the true picture ….

- Prevalence estimates of pfhrp2/3 deletions and mapped the data by country
- **denominator** was all *P. falciparum-positive* samples testing positive by microscopy* and confirmed positive by species-specific polymerase chain reaction testing (PCR)
- 38 publications; 55 studies from 32 countries (01/10-08/19)
- Small sample sizes, heterogeneity in populations, lab methods and estimated prevalence (0-100%)
- 3(5%) of studies met all quality criteria



Systematic reviews

Prevalence of *Plasmodium falciparum* lacking histidine-rich proteins 2 and 3: a systematic review

Rebecca Thomson,[a] Jonathan B Parr,[b] Qin Cheng,[c] Stella Chenet,[d] Mark Perkins[e] & Jane Cunningham[f]

Objective To calculate prevalence estimates and evaluate the quality of studies reporting *Plasmodium falciparum* lacking histidine-rich proteins 2 and 3, to inform an international response plan.
Methods We searched five online databases, without language restriction, for articles reporting original data on *Plasmodium falciparum*-infected patients with deletions of the *pfhrp2* and/or *pfhrp3* genes (*pfhrp2/3*). We calculated prevalence estimates of *pfhrp2/3* deletions and mapped the data by country. The denominator was all *P. falciparum*-positive samples testing positive by microscopy and confirmed positive by species-specific polymerase chain reaction testing (PCR). If microscopy was not performed, we used the number of samples based on a different diagnostic method or PCR alone. We scored studies for risk of bias and the quality of laboratory methods using a standardized scoring system.
Findings A total of 38 articles reporting 55 studies from 32 countries and one territory worldwide were included in the review. We found considerable heterogeneity in the populations studied, methods used and estimated prevalence of *P. falciparum* parasites with *pfhrp2/3* deletions. The derived prevalence of *pfhrp2* deletions ranged from 0% to 100%, including focal areas in South America and Africa. Only three studies (5%) fulfilled all seven criteria for study quality.
Conclusion The lack of representative surveys or consistency in study design impairs evaluations of the risk of false-negative results in malaria diagnosis due to *pfhrp2/3* deletions. Accurate mapping and strengthened monitoring of the prevalence of *pfhrp2/3* deletions is needed, along with harmonized methods that facilitate comparisons across studies.

Abstracts in عربي, 中文, Français, Русский and Español at the end of each article.

https://www.who.int/bulletin/volumes/98/8/20-250621.pdf?ua=1

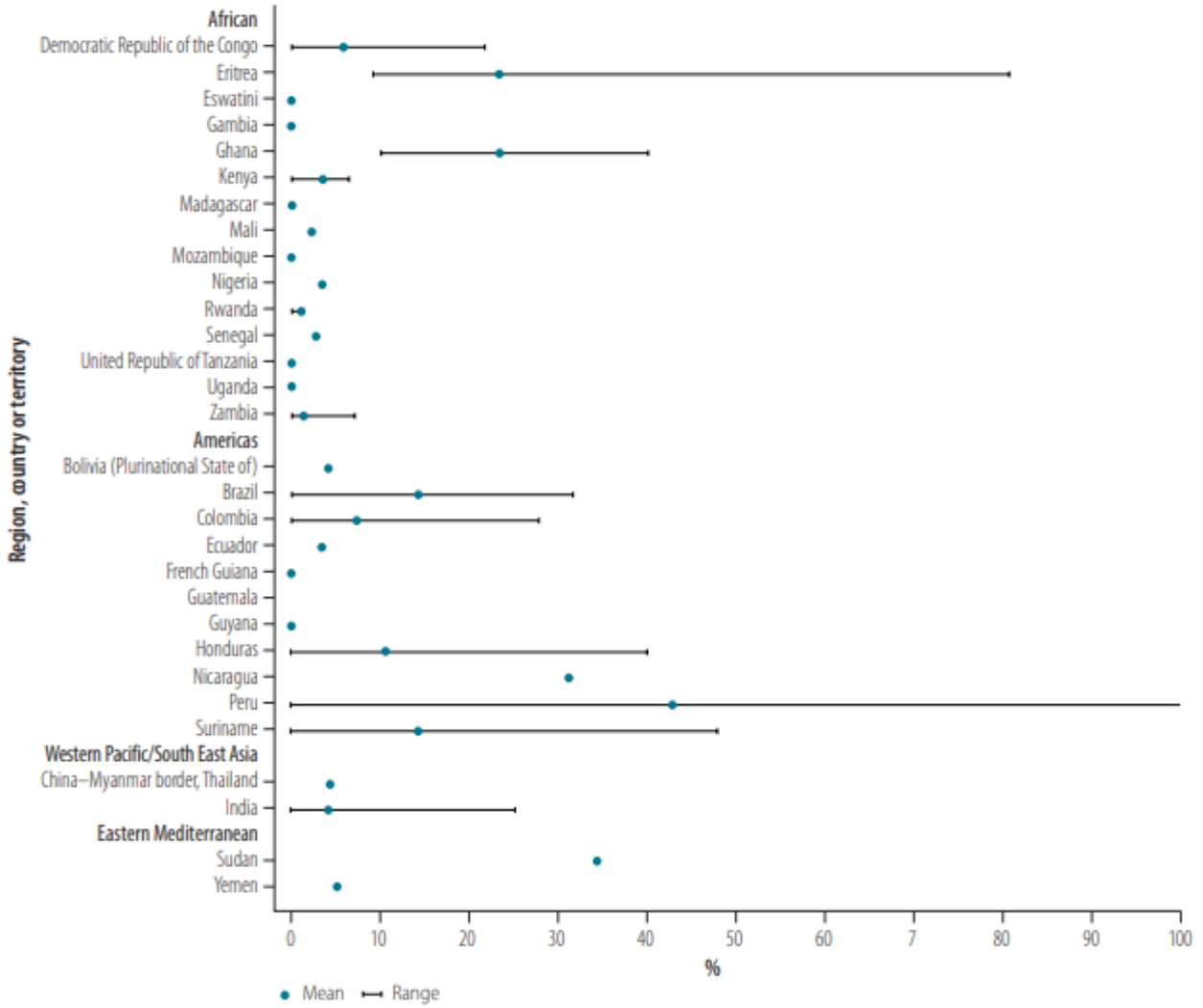
Number of lab-confirmed *pfhrp2/3* deletions

Number of *P.falciparum*-positive samples

*If microscopy was not performed, we used the number of samples based on a different diagnostic method or PCR alone

- Weighted average prevalence of *pfhrp2* gene deletions for each country and the range by study sites.
- The weighted average prevalence ranged from 0% to 43%.
- Average prevalence above 20% was found in Eritrea, Ghana, Nicaragua, Peru and Sudan.

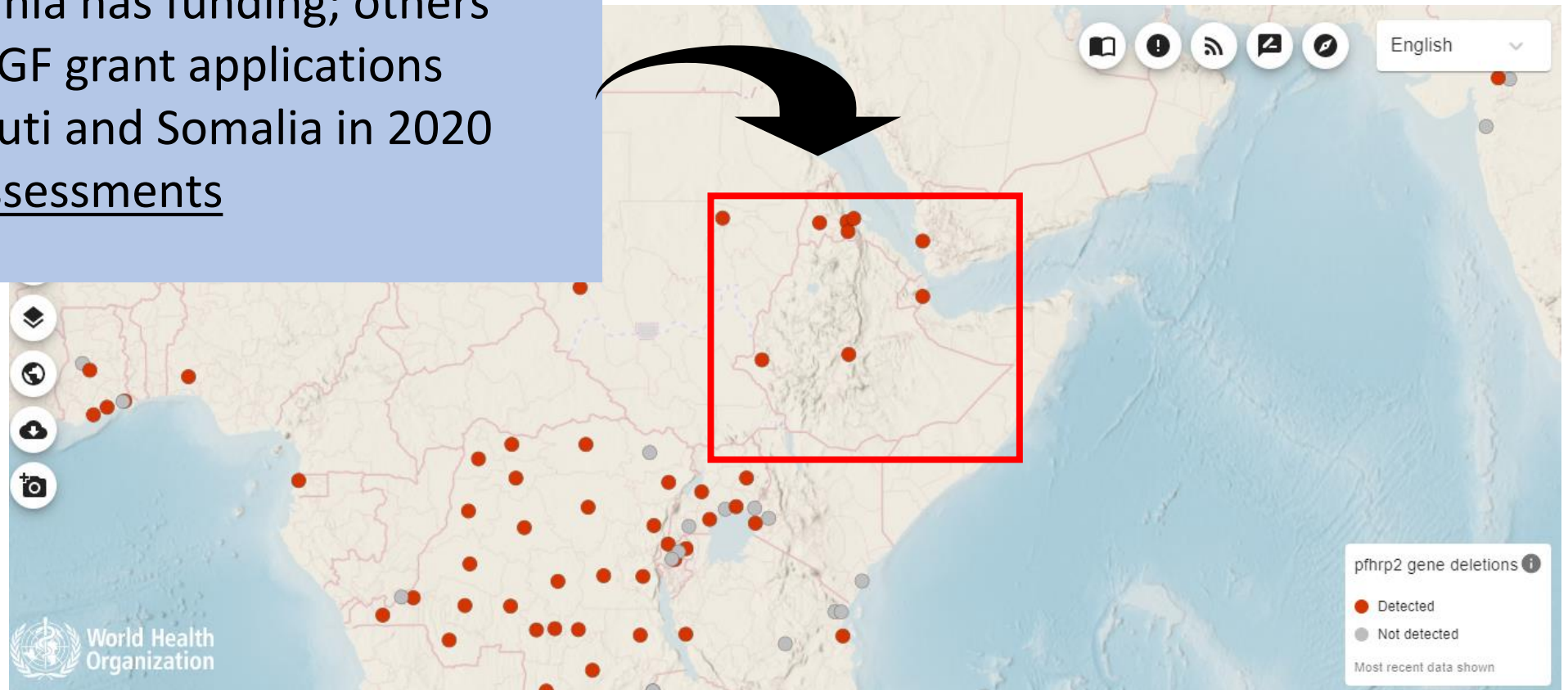
Fig. 3. **Weighted average prevalence estimates for *Plasmodium falciparum pfhrp2* gene deletions among patients tested by country**



Notes: the prevalence of *phfrp2* gene deletions was calculated using the number of *Plasmodium falciparum*-positive samples as the denominator. In countries with multiple studies prevalence estimates were weighted based on the number of patients tested. Countries with only one study have no bar shown. Cases studies and a study from Zambia that tested for *pfhrp2*-leader sequences are not included.

Updates

- Many countries want to conduct surveys **but lack funding**.
- WHO workshop (2019) with 5 countries in SSA – developed country specific protocols an budgets
- In 2020 only Tanzania has funding; others await outcome of GF grant applications
- Signals from Djibouti and Somalia in 2020 prompted rapid assessments

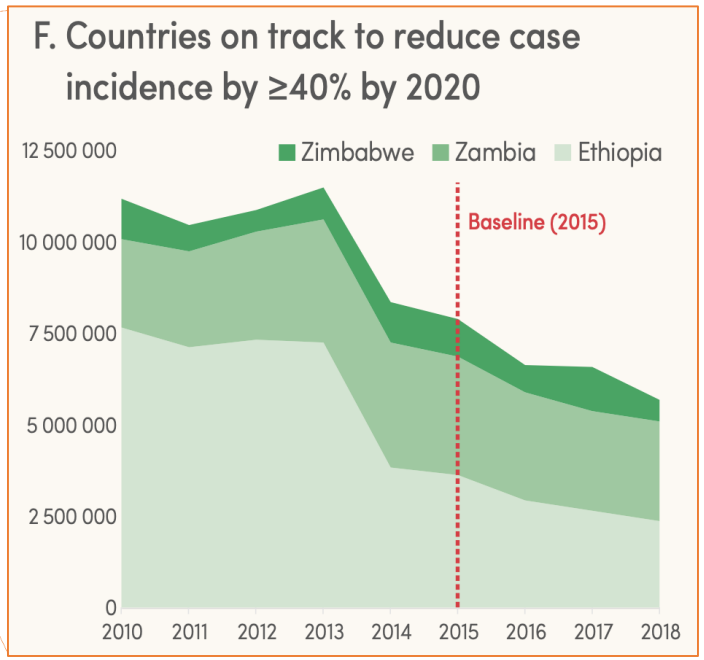


Ethiopia

RESEARCH

Major Threat to Malaria Control Programs by *Plasmodium falciparum* Lacking Histidine-Rich Protein 2, Eritrea

Araia Berhane, Karen Anderson, Selam Mihreteab, Karryn Gresty, Eric Rogier, Salih Mohamed, Filmon Hagos, Ghirmay Embaye, Anderson Chinorumba, Assefash Zehaie, Simone Dowd, Norman C. Waters, Michelle L. Gatton, Venkatachalam Udhayakumar, Qin Cheng, Jane Cunningham



Eritrea

- 62% *pfhrp2-*, RDT- (2016)
- HRP2 RDTs no longer used

Ethiopia

- 2nd most populous country
- Large falciparum malaria burden, but improving
- RDTs introduced 2004, now used for 70% of testing

Berhane A EID 2018; WHO World Malaria Report 2019; Ethiopia Federal MOH 2016

Study design

- Led by the **Ethiopian Public Health Institute**
  - 2017-2018 multi-site, cross-sectional survey
  - To inform malaria diagnostic testing policy

- **First pilot of the WHO protocol**
  - Sampling across diverse regions, districts
  - Use of multiple WHO-prequalified RDTs
  - Molecular typing via reference lab network

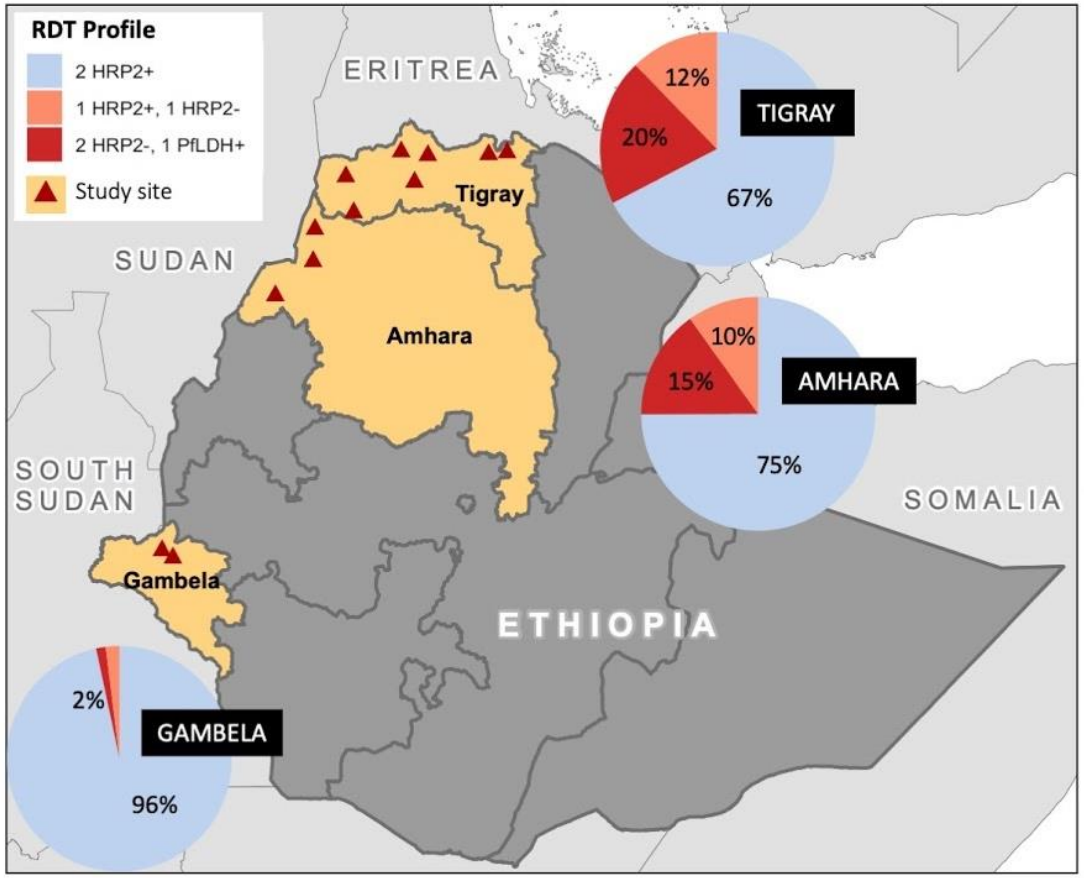
Master protocol for surveillance of *pfhrp2/3* deletions and biobanking to support future research

World Health Organization

https://www.who.int/malaria/publications/atoz/hrp2-deletion-protocol/en/

Suspected deletions were common

- **12,572 subjects** with malaria symptoms
  - 3 regions, 11 districts, 108 facilities
  - 2 WHO-prequalified RDTs:
    - PfLDH RDT: SD Bioline Malaria Ag P.f (**HRP2/pLDH**) (05FK90)
    - Routine RDT: CareStart™ Malaria **HRP2/pLDH** (Pf/Pv) Combo RDT (RMVM-02571)
- **2,707 (22%) with falciparum malaria**
- **354 (13%) with HRP2-,** Pf-pLDH+ bands

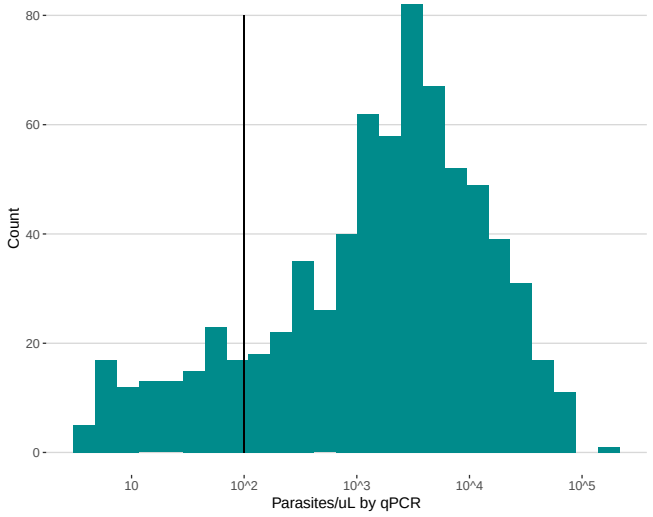


Manuscript submitted for publication

Concordant RDT, PCR, & Luminex calls

926 subjects' DBS for advanced testing

pfldh qPCR → *pfhrp2/3* PCR



↓

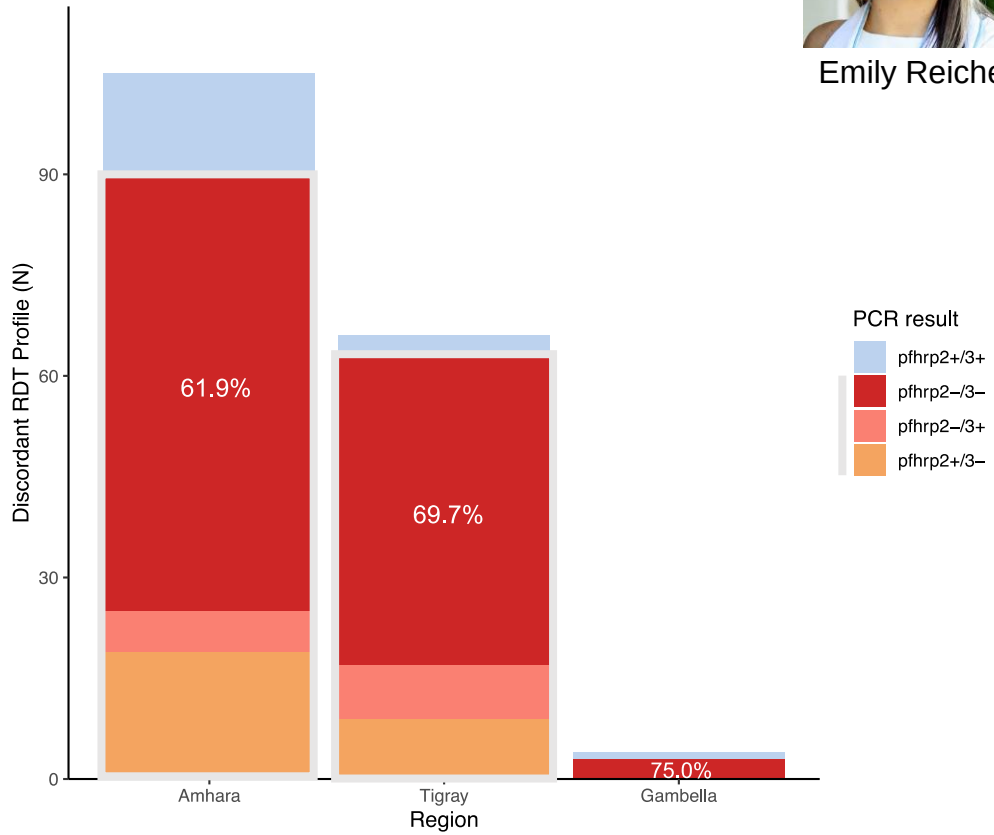
Confirmatory PCR (*pfBtub*)

↓

Putative deletion of *pfhrp2* and/or *pfhrp3*

610 with >100 p/µL
& PCR deletion calls

Emily Reichert



Parr JB, Malar J 2018; Cheng Q Malar J 2014

9.6% (95% CI 8.4-10.9) estimated *pfhrp2*-deleted parasite prevalence among symptomatic falciparum cases overall, meeting WHO criteria for RDT policy change.

Tigray (14.9%; 12.5-17.7)
Amhara (11.5%; 153 9.8-13.4)
Gambella (1.1%; 0.6-2.0).

Published reports of pfhrp2/3 deletions in Ethiopia

PLOS ONE

PLoS One. 2020; 15(11): e0241807.
Published online 2020 Nov 5. doi: 10.1371/journal.pone.0241807

PMCID: PMC7644029
PMID: 33152025

High prevalence and extended deletions in *Plasmodium falciparum hrp2/3* genomic loci in Ethiopia

Lemu Golassa, Conceptualization, Writing – original draft,[1,*] Alebachew Messele, Formal analysis, Writing – original draft,[1] Alfred Amambua-Ngwa, Formal analysis, Writing – review & editing,[2] and Gote Swedberg, Conceptualization, Methodology, Writing – original draft[3]

Takafumi Tsuboi, Editor

Adama town, Oromia – collected in 2015
Malaria suspects – 189 febrile patients; 64 PCR and micro positive (no RDT performed); pfhrp2/3 analysis for 50
100% had deletion pfhrp2 and pfhrp3

Prevalence and Epidemiological Characteristics of Asymptomatic Malaria Based on Ultrasensitive Diagnostics: A Cross-sectional Study

Seble Girma, James Cheaveau, Abu Naser Mohon, Dewdunee Marasinghe, Ruth Legese, Nirujah Balasingam, Adugna Abera, Sindew M Feleke, Lemu Golassa, Dylan R Pillai ✉

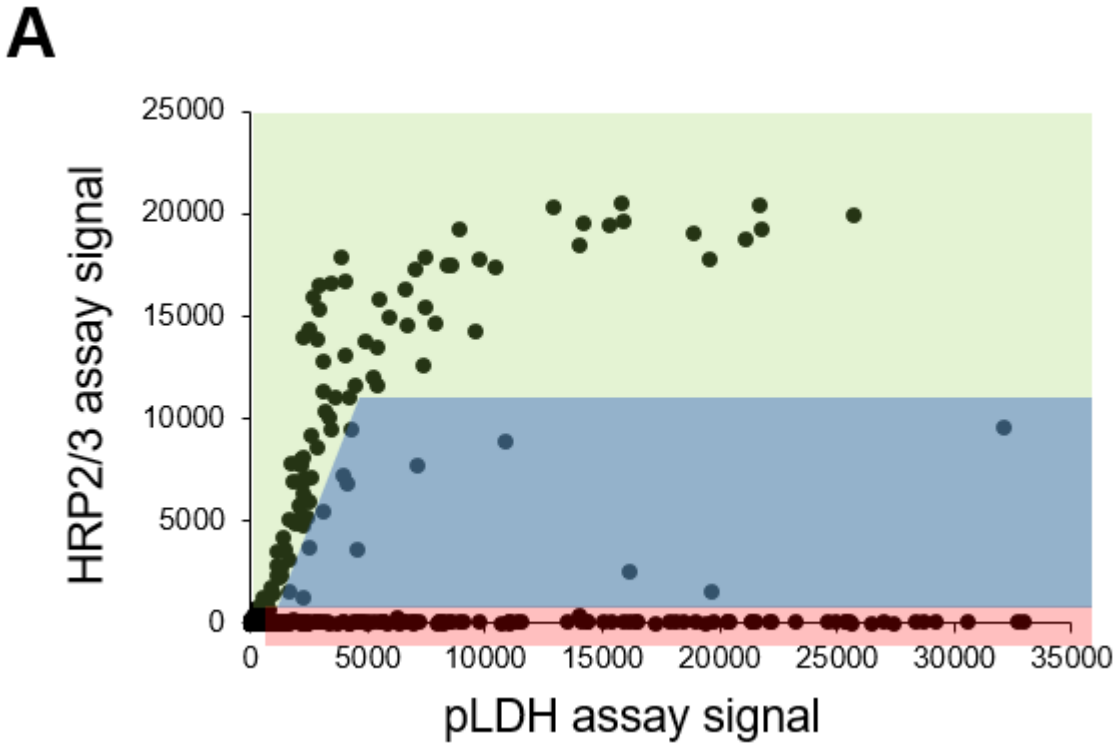
Clinical Infectious Diseases, Volume 69, Issue 6, 15 September 2019, Pages 1003–1010,
https://doi.org/10.1093/cid/ciy1005
Published: 26 November 2018 **Article history ▾**

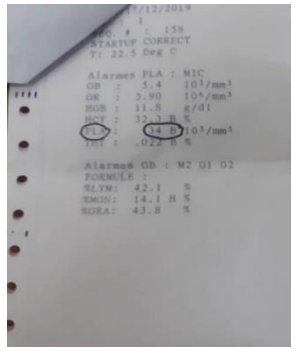
Gambella –published 2018
Asymptomatic – 63 Pf + cases tested;
4.76% pfhrp2 deletions ; pfhrp3 not assessed.

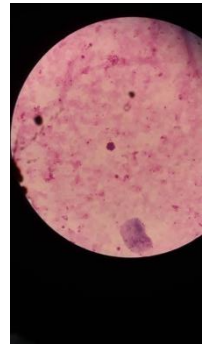
Update: Djibouti – WHO supported rapid assessment

Djibouti – rapid assessment – January 2020

- Reports of false negative RDTs amongst health workers ar lab technicians
- Fever + low platelets + negative HRP2/pv-LDH RDT – microscopy + *P.falciparum*
- Jan- Feb 2020 (Hop. Gen. Peltier) – consecutively prepare DBS from 1002 suspected malaria cases
- May-Sept 2020 (US, CDC)- Luminex (HRP2. pLDH) and PCF
- 189/312 **(60.6%) Pf cases had *pfhrp2* deletions – 98% had both pfhrp2/3 deletions.**







Source: Ministry of Health Djibouti, WHO, US CDC (E.Rogier)

Update : Djibouti – Published report - 2020

Emerg Microbes Infect. 2020; 9(1): 1984–1987.
Published online 2020 Sep 17. doi: 10.1080/22221751.2020.1815590
PMCID: PMC7534257
PMID: 32869688

Misdiagnosis of imported *falciparum* malaria from African areas due to an increased prevalence of *pfhrp2/pfhrp3* gene deletion: the Djibouti case

Xavier Iriart,[a,b,*] Sandie Menard,[b,*] Pamela Chauvin,[b] Hasna S. Mohamed,[c] Elena Charpentier,[a,b] Mohamed A. Mohamed,[c] Antoine Berry,[a,b,†] and Mohamed H. Aboubaker[d,†]

- January-May 2019
- 378 blood samples collected from Djiboutian patients with suspected malaria – Djibouti city

- 20.9% (79/378) samples PCR+ while **HRP2 RDTs negative in 83.5% (66/79) of these samples**.
- Quantitative PCRs targeting the *pfhrp2/pfhrp3* genes confirmed the **absence of both genes for 86.5% of *P. falciparum* strains**

Update: unpublished studies : Somalia and Sudan

Somalia – HRP2 negative RDT + Pf Micro

- Reports of false negative RDTs amongst health workers and lab technicians
- RDTs from suspected cases transported to London, UK
- 17/20 (85%) had pfhrp2 deletions
- 14/20 (70%) had dual pfhrp2 and pfhrp3 deletions
- Rapid assessment survey underway – samples just shipped for molecular analysis

Source: Ministry of Health, WHO, LSHTM (K. Beshir)

Sudan - WHO protocol

- In 2018-2019, WHO provided technical support for WHO Protoc

In two states surveyed 700 Pf cases- only 3 discordant results

- Pfhrp2 deletions confirmed in random selection of samples

Sci Rep. 2020; 10: 12822. PMCID: PMC7393171
Published online 2020 Jul 30. doi: 10.1038/s41598-020-69756-8 PMID: 32733079

***Plasmodium falciparum* isolate with histidine-rich protein 2 gene deletion from Nyala City, Western Sudan**

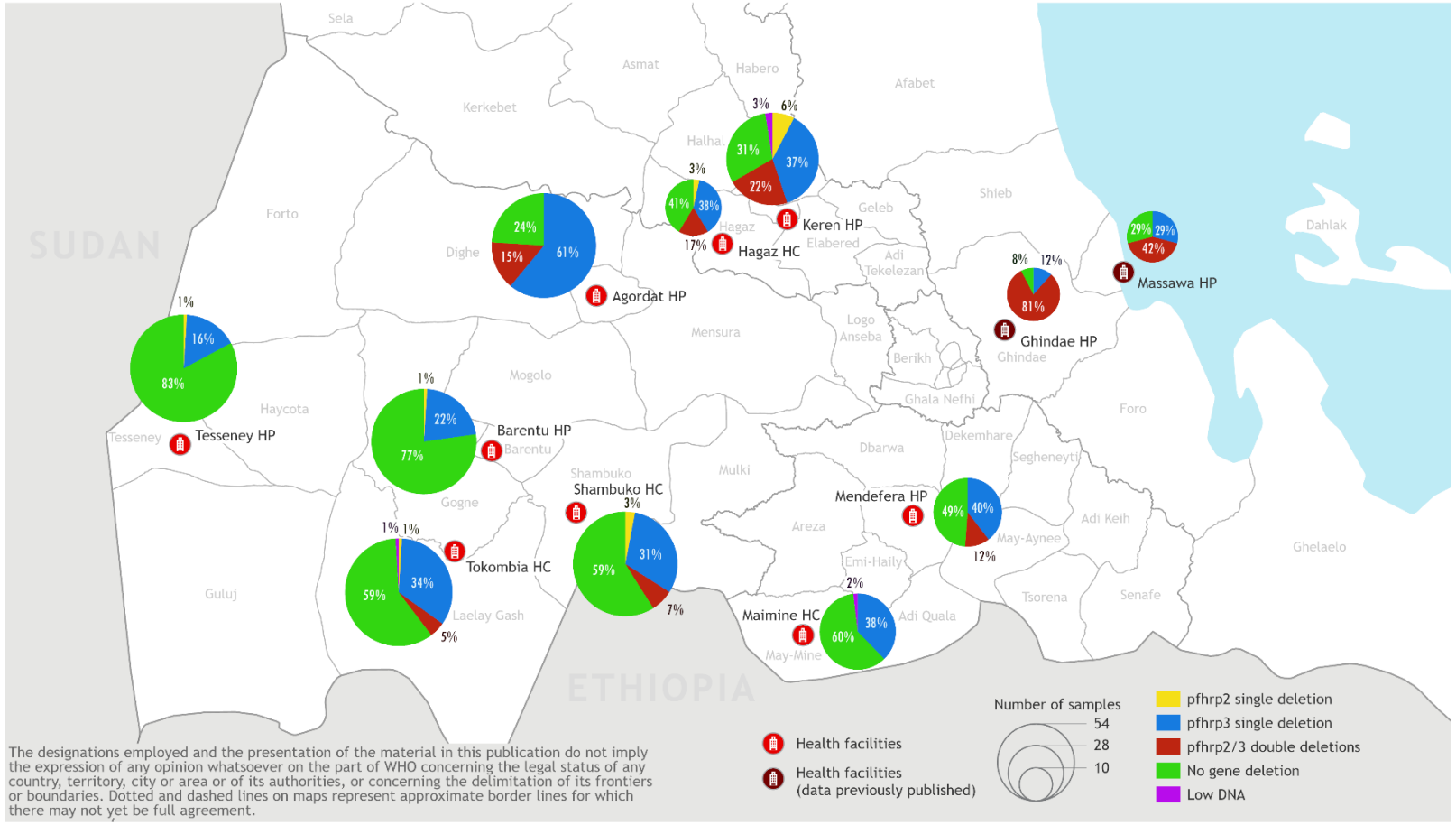
Mohammed A. Boush,[1] Moussa A. Djibrine,[2] Ali Mussa,[3,4,5] Mustafa Talib,[4,5] A. Maki,[6] Abdulrahman Mohammed,[7] Khalid B. Beshir,[8] Zeehaida Mohamed,[9] and Khalid Hajissa[5,9]

- 300 suspected patients screened by microscopy (July 2018- April 2019)
- 113 microscopy positive
- 106/113 (93.8%) detected by HRP2 RDT
- 7 HRP2 negative - PCR confirmation pfhrp2/3
- 1/113 pfhrp2 negative (0.9%)

Update: Unpublished studies – Eritrea

Eritrea – Follow up 2+ yrs post RDT change

- 716 samples from *P. falciparum* confirmed cases from 3 regions
- Mean prevalence of pfhrp2, pfhrp3 and dual deletions – 9.4%, 41.7% and 7.6%, respectively
- Range –dual deletions – 0-21.8%
- **No samples from original sites for direct comparison**
- Deletions persist despite removing the pressure of HRP2 RDTs…….same story in S.America.



Courtesy: MOH, Eritrea and ADFMIDI team, Brisbane Australia

Alternatives

- Eritrea and Djibouti have changed RDTs; Ethiopia planning more surveys …..
  - Eritrea – pan-LDH followed by HRP2/pvLDH RDT for all panLDH +
  - Djibouti – Pf-LDH/Pv-LDH (BioCredit Malaria Ag)

  - WHO-CDC adhoc Biocredit product assessments (2018) and data from other groups eg PATH confirms good performance

Product name	Product code(s)	Manufacturer name	Dossier review	On-site inspection	Laboratory evaluation
BIOCREDIT Malaria Ag Pf (pLDH)	C14RHG25 and C14RHH25	RapiGen Inc.	R		◆
BIOCREDIT Malaria Ag Pf (pLDH/ HRP Ⅱ)	C13RHG25 and C13RHH25	RapiGen Inc.	R		◆
BIOCREDIT Malaria Ag Pf/Pv (pLDH/pLDH)	C61RHG25 and C61RHH25	RapiGen Inc.	R		◆

R	🗁	◆	F	S
information requested from manufacturer	in process	stage complete	follow-up amendments	scheduled; date confirmed

Please note: these tables are updated regularly; while every attempt is made to provide current data, the most recent information might not be reflected. This table is intended only as an update on progress and does not reflect a final decision on prequalification. This table should not be used to inform procurement. Information may not yet be reflected here.

Last update: 2 October 2020

http://www.who.int/diagnostics_laboratory/pq_status/en/index.html

Supply security risk
Elevated price

Conclusions

- Several countries in the Horn of Africa region including Ethiopia have indisputably high prevalence of *pfhrp2* deletions including dual deletions of *pfhrp2 and pfhrp3*
- Eritrea and Djibouti have changed RDTs – pan-LDH and pf-LDH, based on WHO guidance and on findings in a limited geographical area.
- Ethiopia received signals of a problem based on high prevalence of discordant RDTs in 2018 …… 2021 still using HRP2/pv-LDH combo tests which have 0% detection of dual *pfhrp2 and pfhrp3* parasites.
- With continued HRP2 RDT pressure expect problem is ongoing getting worse
- An alternative combo test that does rely on HRP2 is available – in PQ pipeline and GF ERPD approved

Question for MPAC

- What is the best response for countries in the horn of Africa that are still using HRP2 based diagnostics ?
  - More surveillance …how much ?
  - Pre-emptive switch to non-HRP2 based diagnostics for the entire region ?

- In some cases we're identifying the problem after there is already a crisis – how can we bolster support and funding for representative surveys that can guide policy ?
- The 'response plan' calls for research - need a champion ?
  - Factors driving evolution and spread – models suggest high potential for spread when diagnosis relies of HRP2 RDTs
  - Methods to simplify detection
  - New biomarkers

Acknowledgements

- **University of North Carolina**
  - Jonathan Parr
  - Emily Reihart
- **London School Hygiene and Tropical Medicine**
  - Khalid Beshir
- **Australian Defence Force Malaria and Infectious Disease Institute**
  - Qin Cheng
  - Karen Anderson
- **Ethiopian Public Health Institute**
  - Sindew Mekasha
- **US CDC**
  - Eric Rogier
- Ministries of Health, Sudan, Eritrea, Djibouti, Somalia

WHO Regional Malaria Colleagues

- Ghasem Zamani, EMRO
- Jamal Amran, Somalia CO
- Anderson Chinorumba , WHO IST SEA

Malaria Policy Advisory Group Meeting

13—15 April 2021, Geneva, Switzerland
Background document for Session 3

WHO technical consultation on the burden of and response to malaria in urban areas

Strategic Information for Response Unit,
WHO Global Malaria Programme. Geneva, Switzerland

1. Introduction

In the period 2000 to 2030, the world's urban population is expected to increase from 2.7 billion to 5.1 billion, accounting for 60% of the total population *(1)*. This rapidly increasing urbanization has been recognized as a major development, social and health concern, leading to the 2016 launch of the United Nations' (UN) New Urban Agenda as part of the 2030 Agenda for Sustainable Development *(2)*. The World Health Organization's (WHO) Strategic Advisory Group for malaria eradication (SAGme) has identified rapid urban population growth as one of the key megatrends influencing the vision of a malaria-free world *(3)*.

Among the fastest growing regions is sub-Saharan Africa (SSA), which also accounts for over 94% of the current global burden of malaria *(4)*. In this region, the proportion of the population living in urban areas increased from 31% (457 million) to 47% (680 million) between 2000 and 2020. By 2050, 58% of the population in SSA will be urban. In the 10 SSA countries identified for a coordinated global response under the "High burden to high impact" (HBHI) approach, 44% of the population (244 million) was urban in 2020 – a percentage that is projected to rise to 50% (363 million) by 2030. The urban malaria problem is therefore not simply a medium- to long-term concern, but one that needs urgent attention now.

It is expected that well planned urbanization will help to reduce malaria transmission through the destruction of mosquito breeding sites, improved housing, increased living standards, and expanded access to health care *(3)*. However, urbanization in malaria-endemic countries often comes with risks, as large-scale rural to urban migration results in the expansion of unplanned settlements and increased socioeconomic inequity, especially in peri-urban areas and urban slums. These developments can lead to the adaptation of *Anopheles gambiae* s.l. to polluted waters *(5)* and high risk of invasion by *An. stephensi* in certain areas *(6)*. In urban areas, a large fraction of the population seeks malaria treatment in the private sector, potentially receiving substandard care, especially in the uncontrolled informal sector.

However, WHO does not presently have recommendations and implementation guidance specific to urban malaria contexts. In fact, the majority of the evidence underpinning current WHO malaria prevention recommendations relies on efficacy data from rural malaria-endemic settings. Consequently, most countries implement similar interventions in both urban and rural settings, despite important differences in the transmission dynamics and environmental, behavioural, socioeconomic and care-seeking determinants. Furthermore, municipalities often deploy interventions in urban areas that are not recommended by WHO, such as space-spraying, in reducing malaria. Insecticide-treated nets (ITNs) are still widely distributed in African cities, despite little evidence of their efficacy and effectiveness in urban areas, and some data showing that use among those who own nets is often lower in urban areas than in rural ones. Therefore, clear guidance on malaria control in an increasingly complex urban health dynamic is urgently needed. To this end, the

WHO/UCN/GMP/MPAG/2021.03

WHO Global Malaria Programme (GMP) will convene a technical consultation on urban malaria to discuss various themes related to urban malaria in order to develop a global urban malaria control strategy.

General objectives

1. Develop a WHO framework for the response to malaria in urban areas to address the increasing urban population growth and evolving malaria transmission dynamics in malaria-endemic countries.

Specific objectives

1. Document the current practices and lessons learned in the response to urban malaria across WHO regions.
2. Identify effective interventions suitable for reducing the malaria burden and eliminating it in urban settings.
3. Propose methods for urban malaria risk characterization and microstratification to inform targeting of the malaria response.
4. Define urban malaria research priorities and explore issues related to study designs.

2. Proposed guiding questions and context

2.1 How do we define urbanization?

Several methodological approaches have been used to define urban areas and measure urban growth *(7)*. Population size and density, types of housing, infrastructure, economic activities, levels of connectivity and mobility, and national administrative governance all inform the designation of an area as urban. Of the 228 countries or territories for which the UN has assembled population data, more than half use administrative criteria to define urban residents – for example, those dwelling in national or regional capitals or major economic hubs. Most of the others use population size, density or economic characteristics *(1)*. Some have no set criteria.

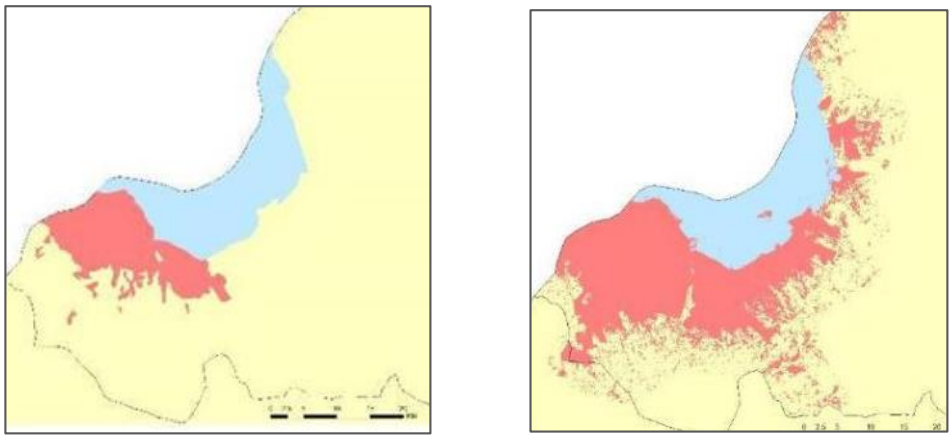
Practically, it is difficult to establish a universal method for defining urban areas so as to reliably compare urban growth within and between countries. However, the absence of such a method poses challenges in defining urban health as a concept and characterizing its unique determinants. This also applies to defining malaria risk and developing urban-specific malaria control recommendations. Some areas defined as urban may in fact have transmission and disease risk characteristics and determinants that are similar to those of rural areas and thus could benefit from similar interventions. Others have unique urban characteristics and may require a different set of interventions.

Increasingly, remote sensing techniques and high-resolution satellite imagery have been used to define urban extents (Fig. 1), based on the principle that urban is a concept of space and not of people *(7, 8)*. Although this is a useful approach for delineating urban geographies, it is crucial to have an understanding of the other factors (demographic, cultural, social, economic, climatic, political, governance, systems and epidemiological) to fully conceptualize urban health within these geographies. It may be that the distinction between urban and rural is not a dichotomy, but rather two ends of a continuum, wherein the transition is defined by degrees of urbanity – a definition that may vary depending on the issues at hand and the relevant conceptual framework to address them. The published literature shows that as the degree of urbanity increases, the general levels of

socioeconomic and health status increase, while recognizing the deep and chronic inequities within some urban settings.

FIG. 1.
a) Change in spatial extent of Kinshasa between 1969 (129 km^2) and 2013 (619 km^2). Spatial extents defined using satellite imagery, administrative and census data *(8)*; b) an aerial view of modern-day Kinshasa showing the contrast in urban development types (https://www.pinterest.ch/pin/546976317223934176/)

a)



b)

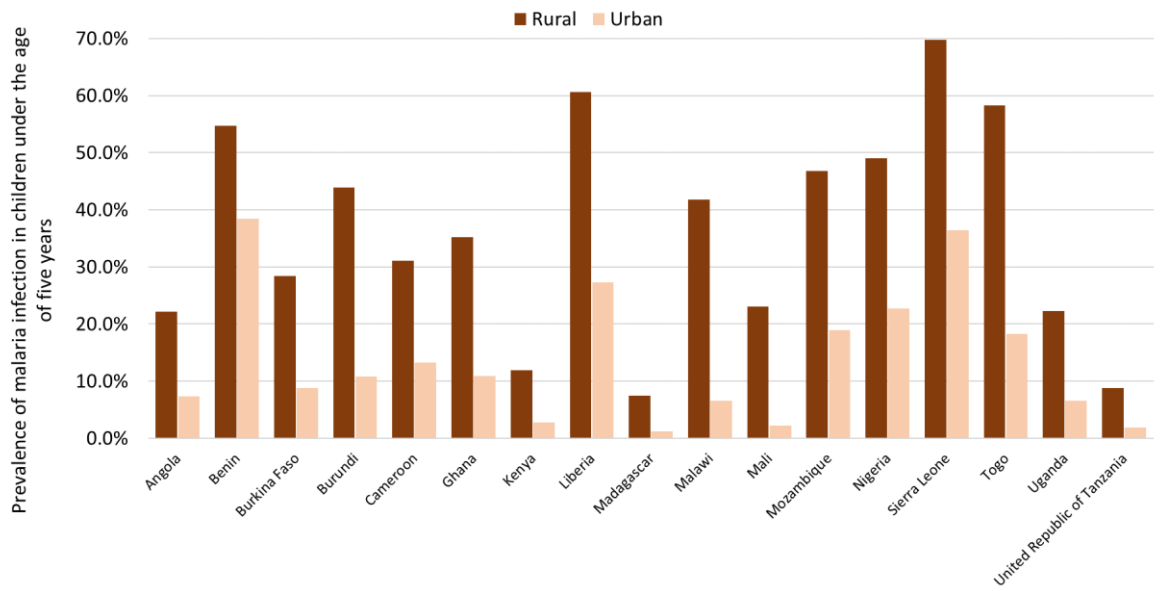
2.2 What drives malaria transmission and disease patterns in urban areas?

Urban malaria risk and burden are heterogeneous over small areas, modified considerably by the built urban environment such as urban agriculture, construction of settlements, roads and water drainage systems, ditches, exposed water channels and open water containers that create common breeding sites for malaria vectors *(9, 10)*. These are often found in urban sectors where people with lower socioeconomic status settle. Overall, however, well planned urban growth has been associated with reduced malaria risk, with evidence that vectors are less numerous per person and the malaria prevalence lower in inner parts of cities than in nearby peri-urban and rural areas *(8)*. Generally, due

to the high level of mobility into and out of urban areas, a large proportion of infections and cases may be acquired outside urban areas.

An analysis of community prevalence surveys implemented in 17 SSA countries since 2015 showed that the prevalence of malaria infection was higher in rural children than in urban children. In 11 countries, prevalence was below 10% in urban areas. In Benin, Liberia, Nigeria, Sierra Leone, Togo and Uganda, more than 20% of children in urban areas who were tested had malaria (Fig. 2). However, further analysis of the data revealed that, even in countries with high urban malaria prevalence, prevalence was much lower in major cities than in other urban areas. For example, in Nigeria, where malaria prevalence in urban areas was about 22% in 2018, Lagos, the largest urban city in the country, had a prevalence of about 3% *(11)*. Overall, trends based on surveys suggest that, for a given level transmission intensity, the more urbanized the setting, the lower the prevalence of malaria.

FIG. 2.
Prevalence of malaria in children under the age of 5 (Source: DHS and MIS surveys)



There are important biases in the available data on infection prevalence and clinical cases in most malaria-endemic urban areas. Standard malariometric surveys do not have large enough sample sizes to reliably capture the granularity of urban malaria risk. Routine case data are usually monthly aggregates without detailed information on age, residence and travel histories, despite the high levels of human population movement to and from urban settings. This makes it difficult to explore age patterns of infection and disease, to understand the rate of local acquisition of infections and to map hotspots. In a study in Nairobi, for example, 22% of patients tested in a facility in an informal settlement were positive for malaria, two-thirds had a history of travel within the month before the examination, and nearly 80% of those who travelled had visited three counties with high malaria transmission *(12)*. Therefore, specially designed urban prevalence surveys and case-based information with travel history are needed to understand the epidemiology of malaria in urban settings. Table 1 describes potential differences between urban and rural settings that will require a differential response to malaria, even when underlying prevalence is similar.

TABLE 1.
Potential differences between urban and rural areas with the same level of parasite prevalence that may elicit a differentiated response to malaria

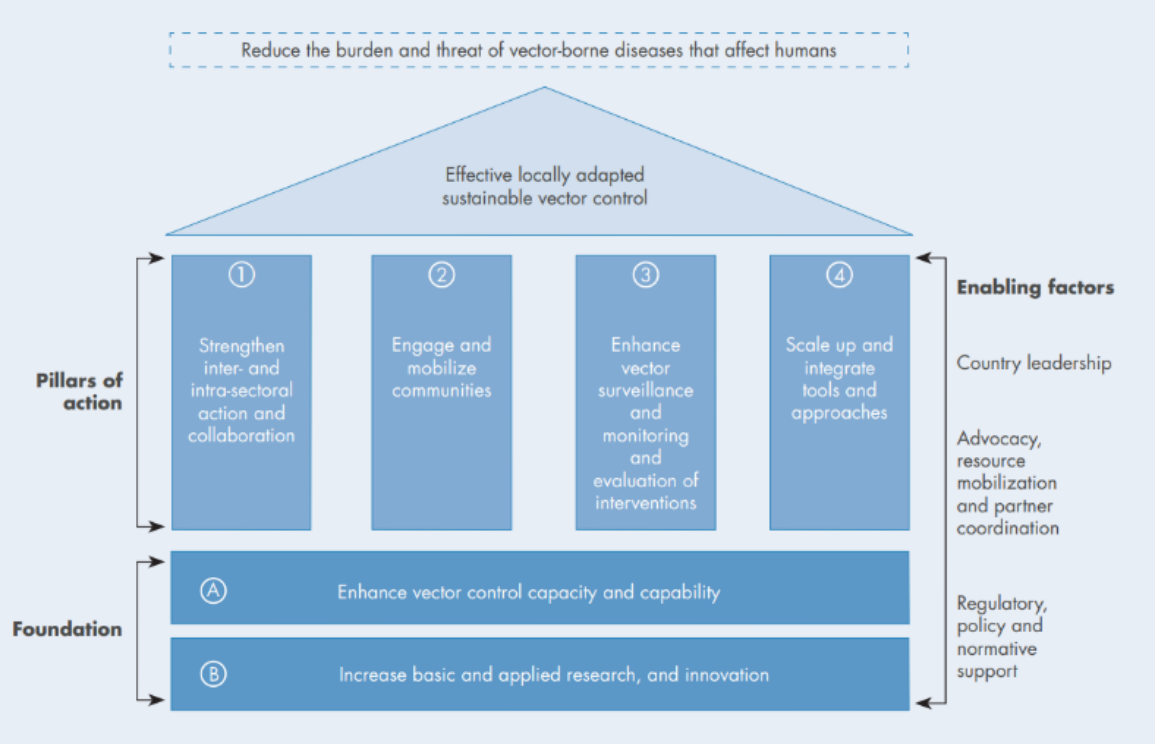
RURAL 5% parasite prevalence, pop. 500 000	URBAN 5% parasite prevalence, pop. 5 000 000
Transmission mainly due to natural ecology	Transmission influenced considerably by environmental modifications and prevalence/incidence influenced by human population movement
Transmission is generalized	Transmission is focal, often higher in peri-urban areas and urban slums with very few areas accounting for most of local infections
Most older children and adults have immunity	Overall population immunity is low
Most infections locally acquired	Large proportion of infections linked to travel to and from higher transmission rural areas
The public health sector is the main source of care for fevers	The private health sector is a major source of care for fevers
High acceptability of IRS and ITNs and use of ITNs	Moderate/low acceptability of IRS and ITNs and use of ITNs
Most housing types allow for high levels of indoor biting	Most housing types reduce indoor biting

2.3 What is the role of ecological and entomological surveillance in understanding urban malaria transmission?

For the reasons explained in preceding sections, and because of the role played by anthropogenic factors, epidemiological data alone may be insufficient to accurately characterize urban malaria risk and determinants in order to mount an effective response. Ecological approaches, including a core entomological component, are likely to be an essential dimension in understanding and mapping malaria risk in urban settings. These approaches are likely to be as relevant to malaria as they are to several other vector-borne diseases (VBDs) that are major causes of ill health in urban populations. These include dengue, Chikungunya, yellow fever and Zika virus diseases, leishmaniasis and lymphatic filariasis.

Vectors such as *Aedes aegypti* and *Culex quinquefasciatus* are highly adaptable to urban settings, as are increasingly, some of the malaria vectors such as *An. gambiae* s.l. *(5)*. With the high levels of human mobility, *An. stephensi*, a highly efficient malaria vector has invaded several cities in the Horn of Africa resulting in outbreaks *(6)*. This vector was prior to 2012 not reported in Africa. Unplanned urban expansion, poor drainage and sanitation, inadequate housing, population movement and climate change are contributing to the rising transmission of VBDs in urban areas. The diversity of the vector ecology in urban areas presents an opportunity to start applying the integrated vector surveillance and control approaches outlined in the WHO Global Vector Control Response (GVCR) strategy (Fig. 3). This will require a significant expansion of VBD entomological surveillance in urban settings, which is currently implemented in an uncoordinated way with few active surveillance sites.

FIG. 3.
WHO Global Vector Control Response Framework *(13)*

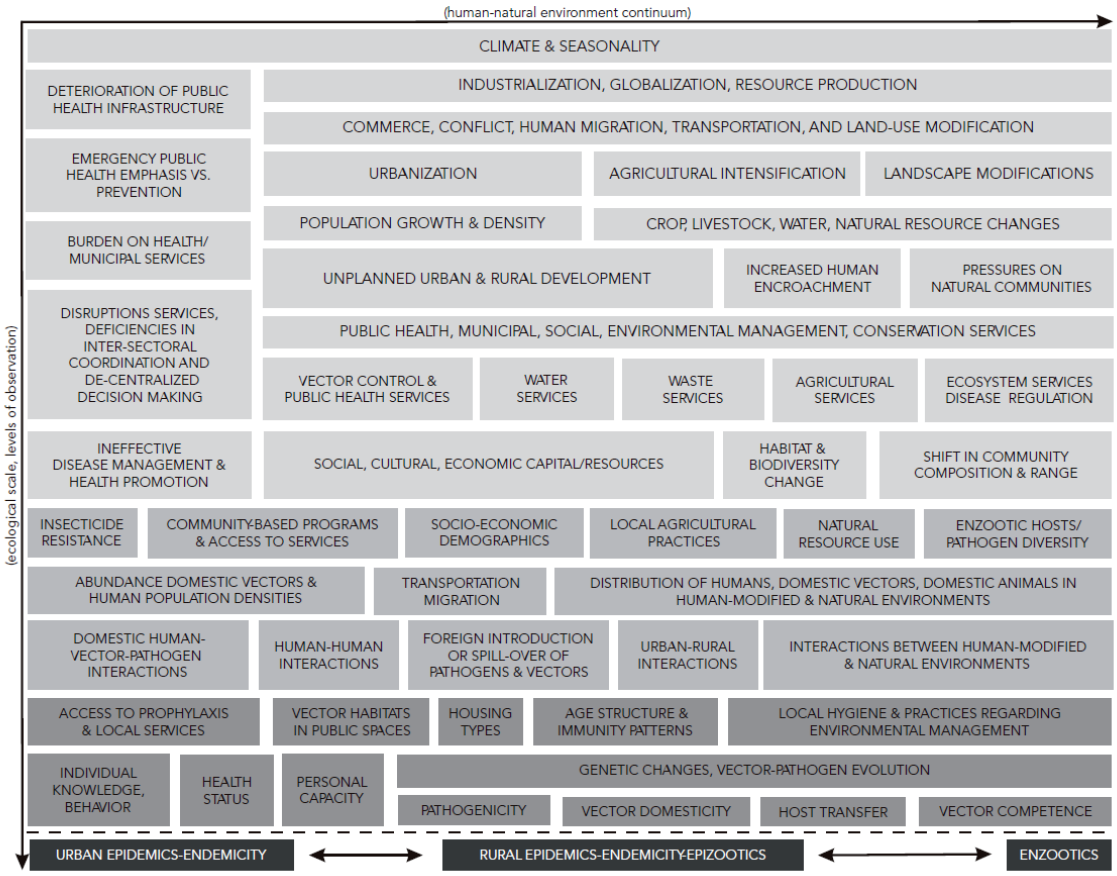


2.4 What information do we need and what stratification methods should we use to tailor malaria interventions to urban settings?

Appropriate intervention mixes in urban areas will be defined by three main requirements: i) the spatial scale will be smaller and granular data will be essential; ii) the approach must combine ecological (including social, environmental, economic and system issues), entomological and epidemiological factors; and iii) decisions will need to be made on the use of approaches for which there are no WHO recommendations and for which the available evidence is of low quality.

High-resolution satellite imagery, multisector urban data and advanced geospatial analysis methods provide exceptional opportunities to implement urban microstratification and intervention tailoring. Existing conceptual frameworks derived from the ecological, geographic, agricultural and cartographic disciplines can be adapted to inform the characterization of the urban environment. An example of a framework that addresses potential ecological interactions related to VBDs *(14)* is presented in Fig. 4. Simplified adaptation of such a framework could help guide the process of urban microstratification and intervention tailoring.

FIG. 4.
Spectrum of ecological interactions associated with VBD transmission *(14)*



2.5 What are the current approaches for malaria prevention and treatment in urban settings and which ones have a WHO policy recommendation?

There are no WHO-recommended malaria interventions that are specific to urban settings. In most urban areas, the interventions implemented are the same as those in rural areas. In other urban areas, some of the malaria control interventions used are not recommended by WHO (Table 2), as the necessary evidence may not be available, despite observational data showing that some of them are strongly associated with reductions in malaria risk *(15, 16)*. Although there is a universal need for access to prompt diagnosis and treatment, several studies have shown that many urban patients who rely on treatment in the private sector may be receiving substandard medicines. In the absence of trial data, WHO relies on other studies of lower quality evidence, such as observational studies and mathematical modelling techniques. Such evidence could be explored through WHO guidance review processes to develop relevant recommendation for the use of these interventions in urban settings.

TABLE 2.
A summary of direct and indirect approaches to malaria prevention and treatment in urban settings

TYPE	INTERVENTION	HAS WHO MALARIA POLICY RECOMMENDATION?
Environmental modification	• Improving drainage	No
	• Draining swamps	No
	• Dredging to increase water flow	No
	• Making embankments	No
	• Land reclamation	No
	• Deforestation/afforestation	No
	• Flood control	No
	• Improved sanitation including better water storage and provision and good maintenance of piped water	No
	• General infrastructure development – e.g., construction of roads	No
Social/ preventive	• House/window screening	No
	• Improved housing	No
	• House inspections to identify and remove breeding sites	No
Vector control	• Larviciding	Yes
	• Aerial spraying of insecticides	No
	• Indoor residual spraying with insecticides	Yes
	• Insecticide-treated bed nets	Yes
	• Entomological surveillance of breeding sites	Yes
	• Monitoring of insecticide resistance	Yes
Antimalarial drugs	• Early diagnosis and treatment by health care providers	Yes
	• Prophylaxis	No
	• Intermittent prevention in children	No
	• Intermittent prevention in pregnant women	Yes (in some settings)
	• Self-treatment at home/informal sector to increase access	Yes (in some settings)
	• Mass screen and treat	No

2.6 How do we ensure that urban governance and leadership in malaria-endemic countries prioritize malaria as part of broader urban development and health systems?

The political and fiscal empowerment of urban governments, their levels of efficiency and accountability, the extent of technological and infrastructure development, and the underlying socioeconomic status and equity are all critical to the health of urban populations. Urban governance is quickly evolving as urban populations increase rapidly, their demographic structure changes, and technological uptake improves; at the same time, the challenging needs of large populations in confined spaces must be dealt with *(17)*. A global Urban Governance Survey conducted by a network convened by UN Habitat considered a range of governance issues. A summary of the relevance of these issues is shown in Table 3.

In many malaria-endemic countries, urban governance is weak, even in decentralized systems where subnational entities are responsible for health policy implementation and resource decisions. The coordination and stewardship of national malaria programmes has limited influence over urban pest control or drug outlets dominating the informal private sector. This limits the scale and focus of the malaria interventions that urban governments fund and/or implement. To encourage governments to increase and improve the efficiency of their domestic expenditures on malaria, urban malaria control is likely to be an attractive opportunity for governments, given the considerable role of major cities in national politics and economy. A good understanding of the governance landscape must therefore underpin the practical adaptation of global urban malaria control policies.

TABLE 3.
Results of the global urban governance survey 2014–2015 *(17)*; number of cities that answered *very relevant* or *highly relevant* to the question, "To what extent are the following issues challenges to governing your city?"

Ranking	Urban governance challenge category	Number of cities, *n* (%)
1	Insufficient public budgets	28 (50)
2	Politicization of local issues	21 (38)
3	Interdependence of policy issues	21 (38)
4	Inflexible bureaucracies/rigid rules	20 (36)
5	Lack of municipal autonomy	17 (30)
6	Overlapping responsibilities	17 (30)
7	Working across different tiers of government	17 (30)
8	Access to useful information	16 (29)
9	Lack of respect for laws and regulations	15 (27)
10	Lack of capacity to enforce laws and regulations	15 (27)
11	Lack of skills in local government	14 (25)
12	Uncertainty of funding	14 (25)
13	Risks of corruption	13 (23)
14	Limited scope of responsibilities	13 (23)
15	Coordination of different sectors/departments	13 (23)
16	Limited access of citizens to policymaking	11 (20)
17	Lack of interest of citizens on local issues	11 (20)
18	Lack of trust in local government	10 (18)
19	Lack of political stability	8 (14)
20	Underrepresentation of vulnerable groups	6 (11)

Note. Data from LSE Cities, UN-Habitat, and UCLG (2016).

2.7 What opportunities do urban planning and health systems provide in the malaria response?

There has been limited systematic analysis of the relationship between urbanization and urban health in low- and middle-income countries (LMICs), and less so the relationship between urbanization and the control of malaria and other VBDs. Compared to rural residents, urban communities have access to more health workers, financial resources and facilities, higher access to electricity and better supply chain management. High population density facilitates large-scale access to health care facilities and services. However, these settings also have specific vulnerabilities to infectious diseases, as increasing informal settlements, poor housing, air pollution, poor drainage, insecurity, high population mobility and deep inequalities to prevention and care all increase the level of disease transmission, with high population densities increasing the potential risks for large outbreaks.

The New Urban Agenda was adopted at the United Nations Conference on Housing and Sustainable Urban Development (Habitat III) in Quito, Ecuador on 20 October 2016, and endorsed by the General Assembly in December 2016 *(2)*. The Agenda, anchored by the SDGs (Fig. 5), was seen as representing "*a shared vision for a better and more sustainable future – one in which all people have equal rights and access to the benefits and opportunities that cities can offer, and in which the international community reconsiders the urban systems and physical form of our urban spaces to achieve this*".

Although limited in specifics, Member States declared: "*We commit ourselves to fostering healthy societies by promoting access to adequate, inclusive and quality public services, a clean environment, taking into consideration air quality guidelines, including those elaborated by the World Health Organization, and social infrastructure and facilities, such as health-care services, including universal access to sexual and reproductive health-care services to reduce newborn, child and maternal mortality.*"

This Agenda, therefore, provides a platform through which the global malaria community can begin to engage malaria-endemic countries to ensure that control of the disease is firmly part of their broader urban health response.

Fig. 5.
Conceptual framework: Urban health-related SDGs within a Health in All Policies (HiAP) approach in the context of SDG implementation (18)

2.8 What are the key knowledge gaps and priority research questions in malaria transmission dynamics and response?

In 2015, the Special Programme for Research and Training in Tropical Diseases (TDR) commissioned six scoping reviews on the prevention and control of VBDs in urban areas *(19, 20)*. The *VEctor boRne DiseAses Scoping* (VERDAS) reviews identified the following priority research themes:

- interventions
- community and society
- technologies and equity
- ethics

- population mobility
- city responsibility
- transmission and interaction
- surveillance
- collaboration and health services (clinics).

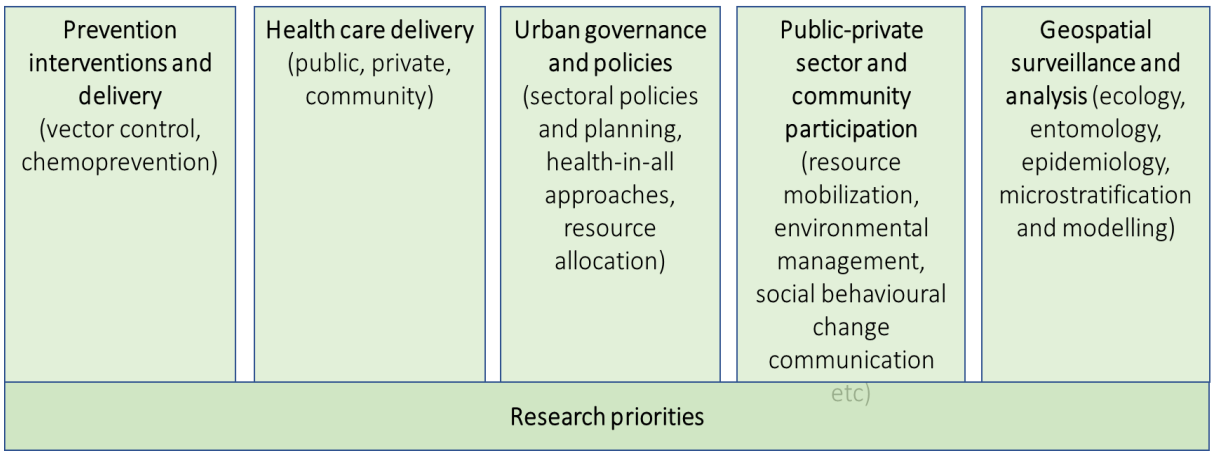
Building on the VERDAS scoping reviews and methodological approach, the technical consultation will aim to define the research priorities for urban malaria transmission and response.

3. Method of work

Almost all of the consultations will be virtual. Key thematic work areas will be identified in the initial convening. Proposed thematic areas include prevention intervention and delivery; health care delivery; urban governance and policies; public-private and community participation and geospatial surveillance and analysis. All thematic areas will also focus on research priorities within their thematic areas. These thematic areas will be discussed during the initial meeting and necessary changes to them will decided on at this time.

A group of 4–5 people, including at least one WHO representative and a rapporteur, will be assigned to each thematic area. A chair will be selected by each thematic group. Thematic groups will use weekly two-hour meetings to develop key topics and content. Each month, WHO will convene a three-hour meeting of all thematic groups for progress updates from each group and feedback from members of the technical consultation.

FIG. 6.
Proposed thematic work areas



4. Proposed membership

Experts in the following areas:

1. Malaria epidemiology
2. Urban malaria entomology, VBD surveillance and vector control
3. Urban environmental health, health care provision (including malaria case management)
4. Urban governance, planning and health systems

5. Urban geospatial mapping and microstratification
6. Multisectoral engagement

Secretariat:

1. WHO GMP
2. WHO Regional representatives
3. WHO selected country representatives
4. WHO environmental and urban health
5. WHO-TDR
6. WHO-NTD

Partners:

1. Funders
2. Collaborating centres
3. RBM Partnership to End Malaria
4. Africa CDC
5. UN Habitat

5. Timelines

The call for membership will be online within the last week of April 2021. Selection of members will be completed by second week of May, at which time the first meeting of the technical consultation will take place. The process will continue until the end of August 2021, with the aim of completing a draft technical strategy by the end of September.

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Technical consultation on urban malaria burden and response

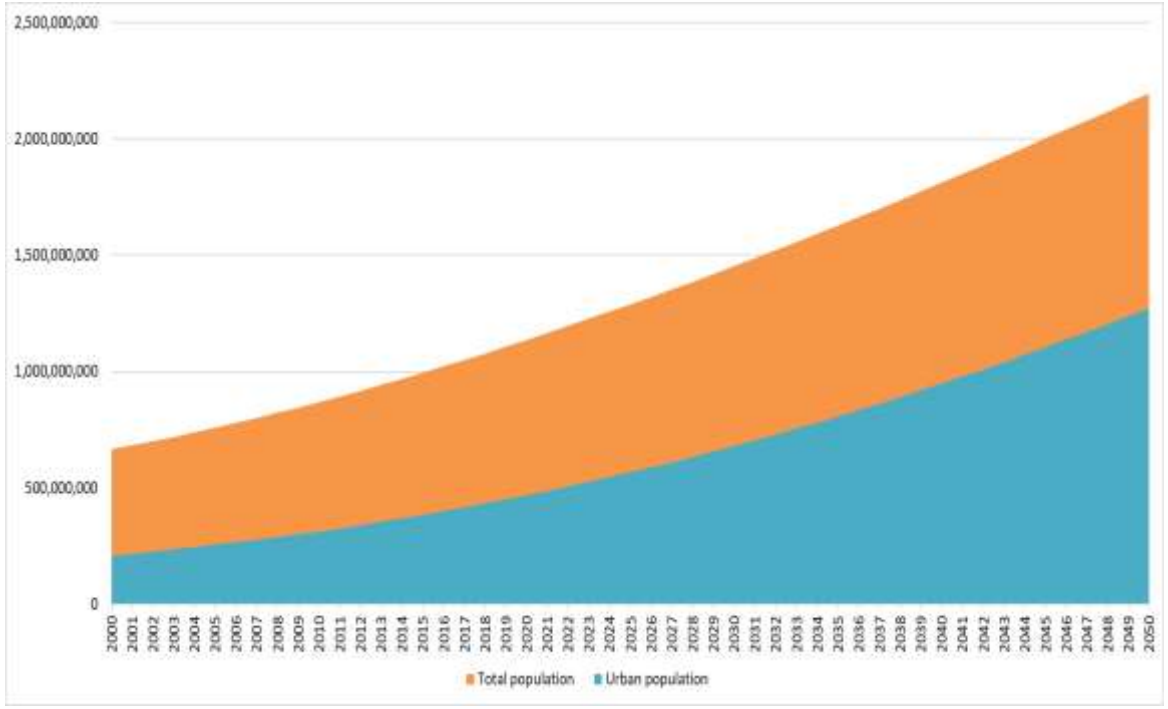
Dr Abdisalan M Noor
Head of Unit
Strategic Information for Response

April 2021
Meeting of the
Malaria Policy Advisory Group

Global Malaria Programme | World Health Organization

Background

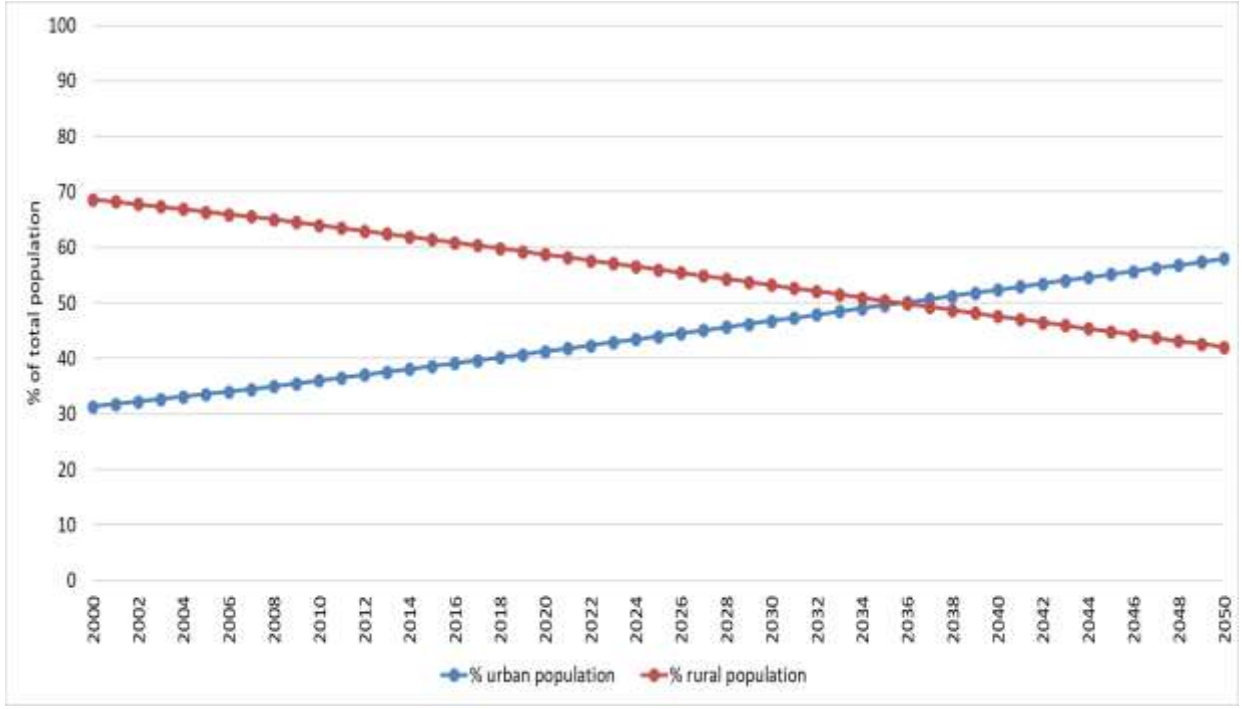
Population count, SSA



In the 10 highest burden countries in SSA, 43% of population already in urban areas in 2020

Rapid Urban Population Growth in malaria endemic countries

% urban, rural, SSA



Background

Potential differences between urban and rural areas the same level (even at the same level of parasite prevalence), may elicit different response to malaria

Rural, **5% parasite prevalence, pop. 500 000**	**Urban,** **5% parasite prevalence, pop. 5 000 000**
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Most infections locally acquired	Large proportion of infections linked to travel to and from higher transmission rural areas
The public health sector is the main source of care for fevers	The private health sector is a major source of care for fevers
High acceptability of IRS and ITNs and use of ITNs	Moderate/low acceptability of IRS and ITNs and use of ITNs
Most housing types allow for high levels of indoor biting	Most housing types reduce indoor biting

General objectives

Develop a WHO framework for the response to malaria in urban areas to address the increasing urban population growth and evolving malaria transmission dynamics in malaria-endemic countries.

Specific objectives

1. Document the current practices and lessons learned in the response to urban malaria across WHO regions.
2. Identify effective interventions suitable for reducing the malaria burden and eliminating it in urban settings.
3. Propose methods for urban malaria risk characterization and microstratification to inform targeting of the malaria response.
4. Define urban malaria research priorities and explore issues related to study designs.

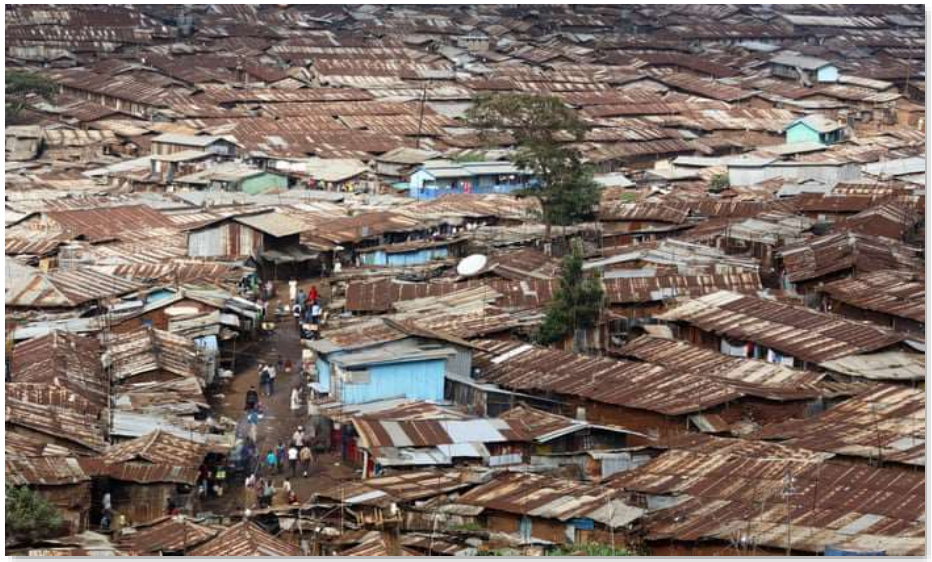
Differences between cities

Mafinga, Tanzania

Dar es Salaam, Tanzania

Differences within cities

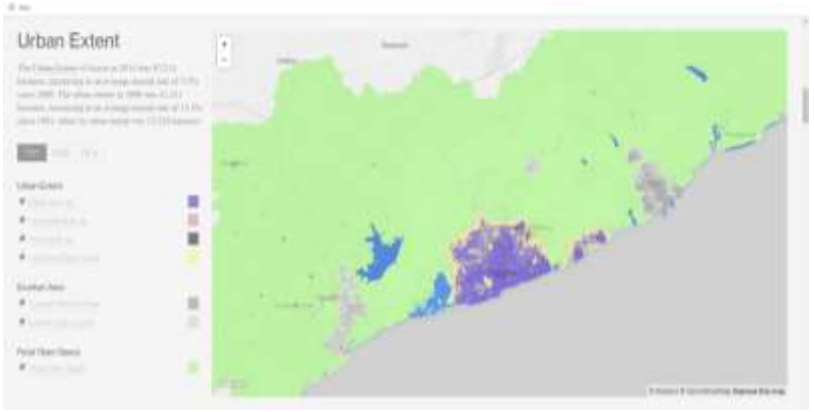
Kibera, Nairobi

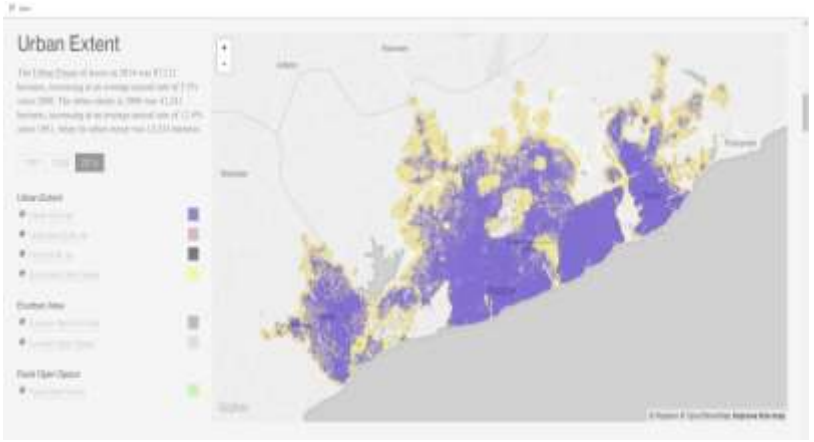


Westlands, Nairobi

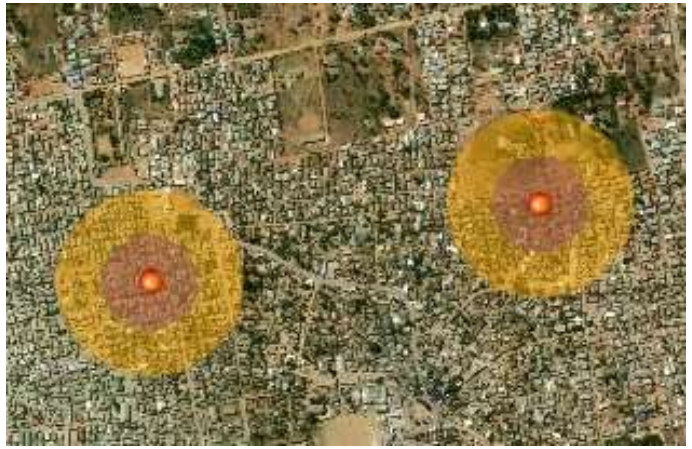
How do we define urbanization?

Accra





High resolution geospatial mapping

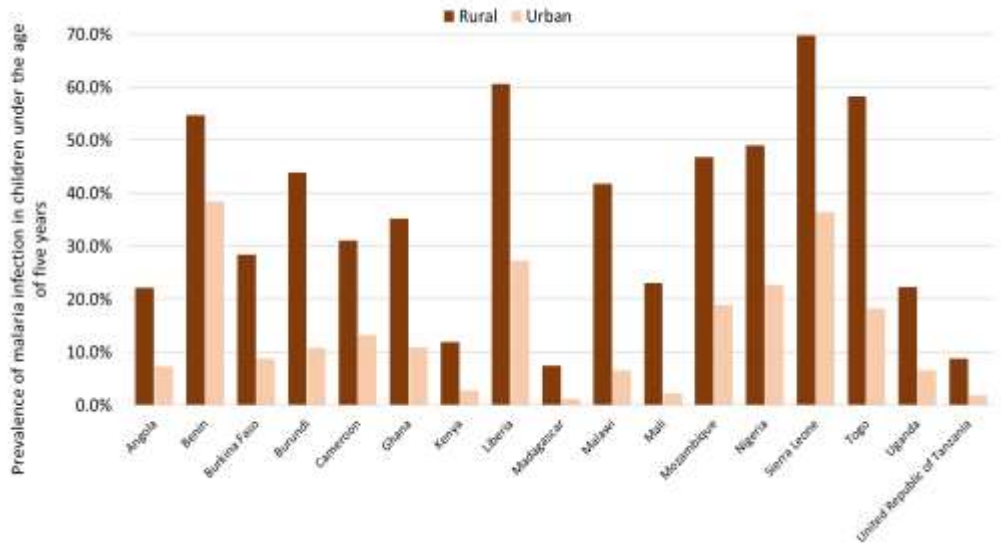


https://grid3.org/news/african-govts-utilise-grid3-data-in-response-to-covid-19

How do we define urbanization?

http://www.atlasofurbanexpansion.org/cities/view/Accra

Parasite prevalence

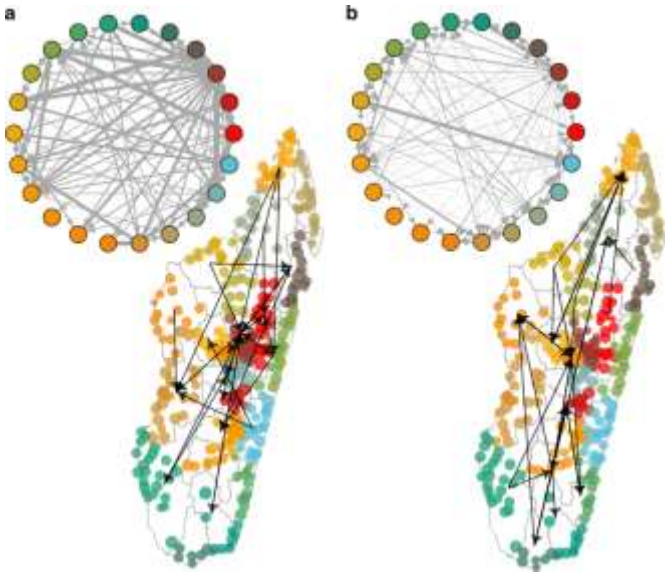


What drives malaria transmission and disease patterns in urban areas?

Ecology of larval habitats

Azrag & Mohammed (2018)

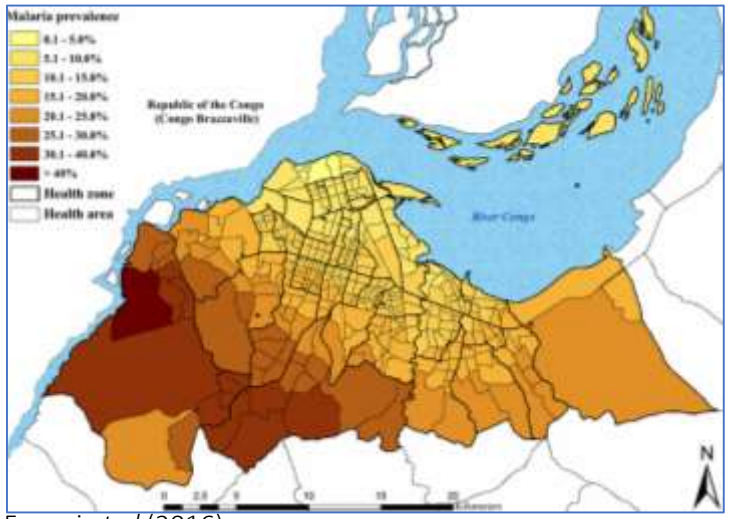
Travel and parasite importation networks



Ihantamalala *et al* (2018)

Parasite prevalence

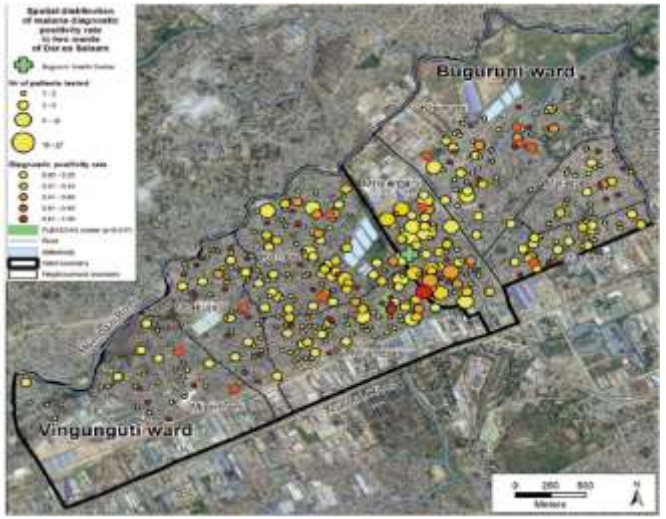
Kinshasa



Ferrari *et al* (2016)

Case incidence

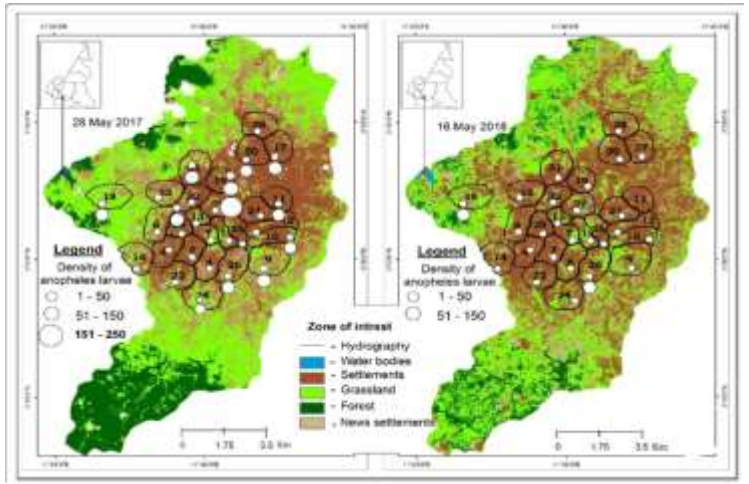
Dar es Salaam



Mlacha *et al* (2017)

Land cover and larval density

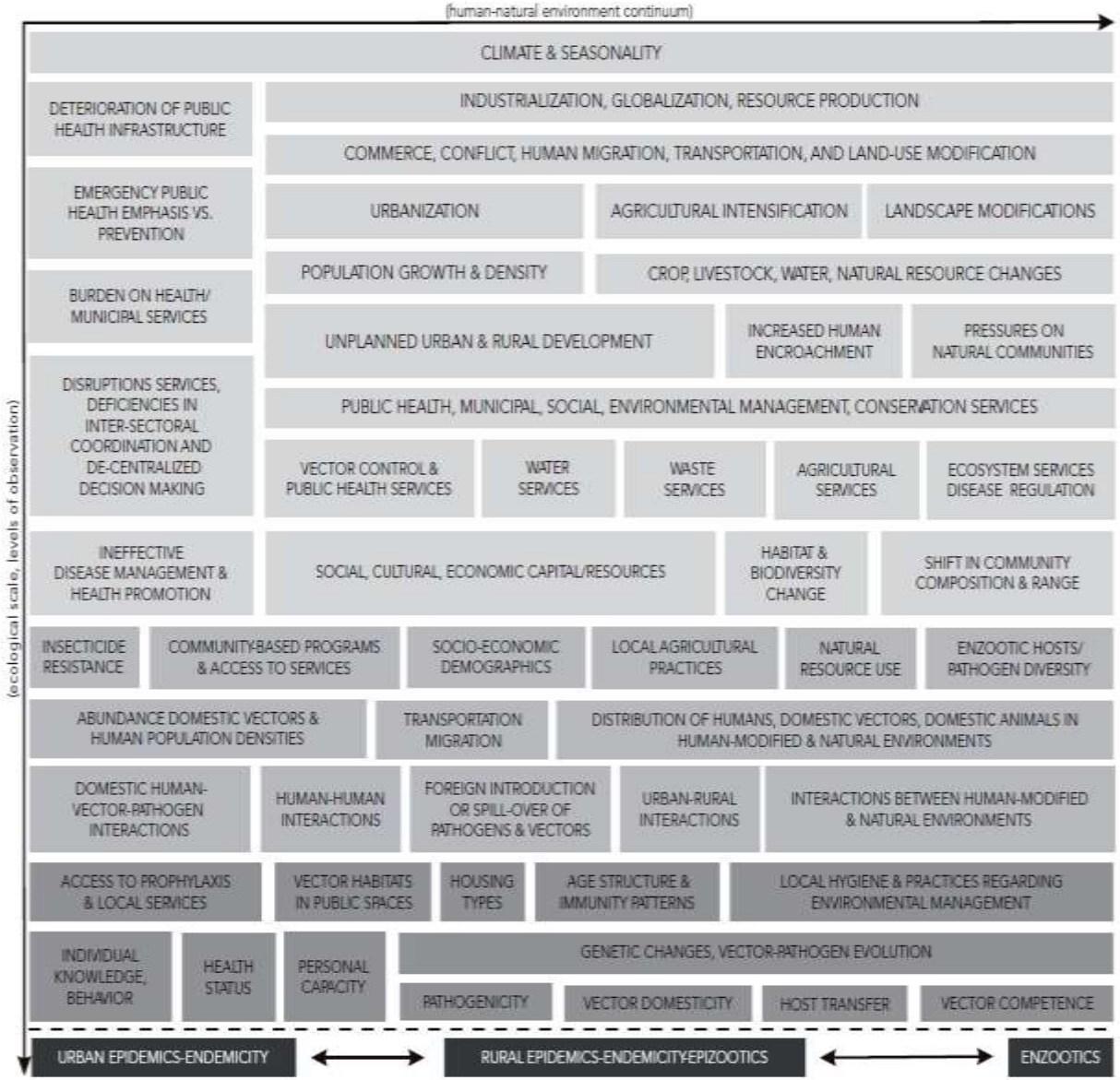
Yaounde



Djamouko-Djonkam *et al* (2019)

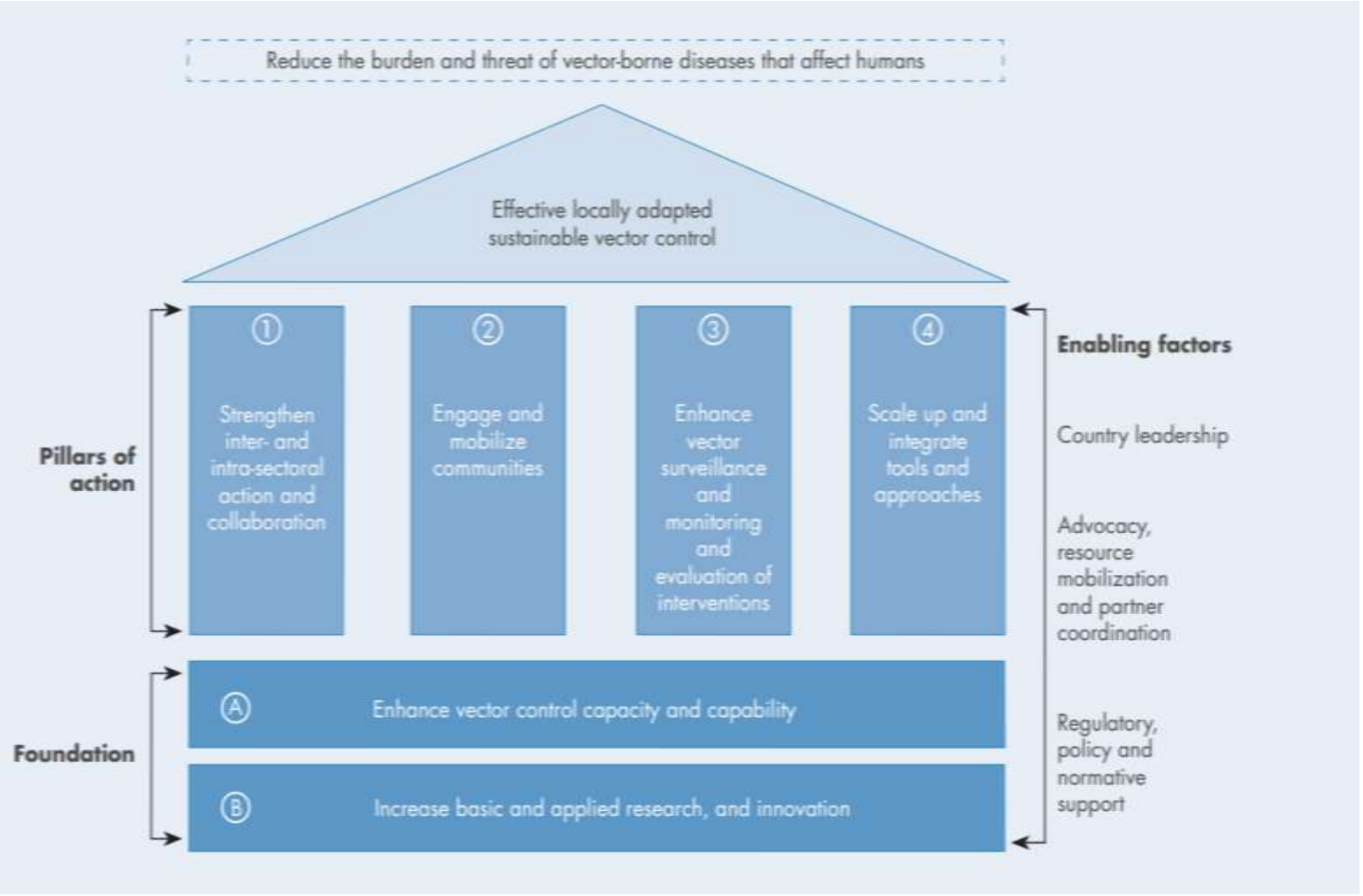
What information do we need and what stratification methods should we use to tailor malaria interventions in urban settings?

Spectrum of ecological interactions associated with VBD transmission



Ellis & Wilcox *et al* (2017)

What information do we need and what stratification methods should we use to tailor malaria interventions in urban settings?



Global Vector Control Response Framework

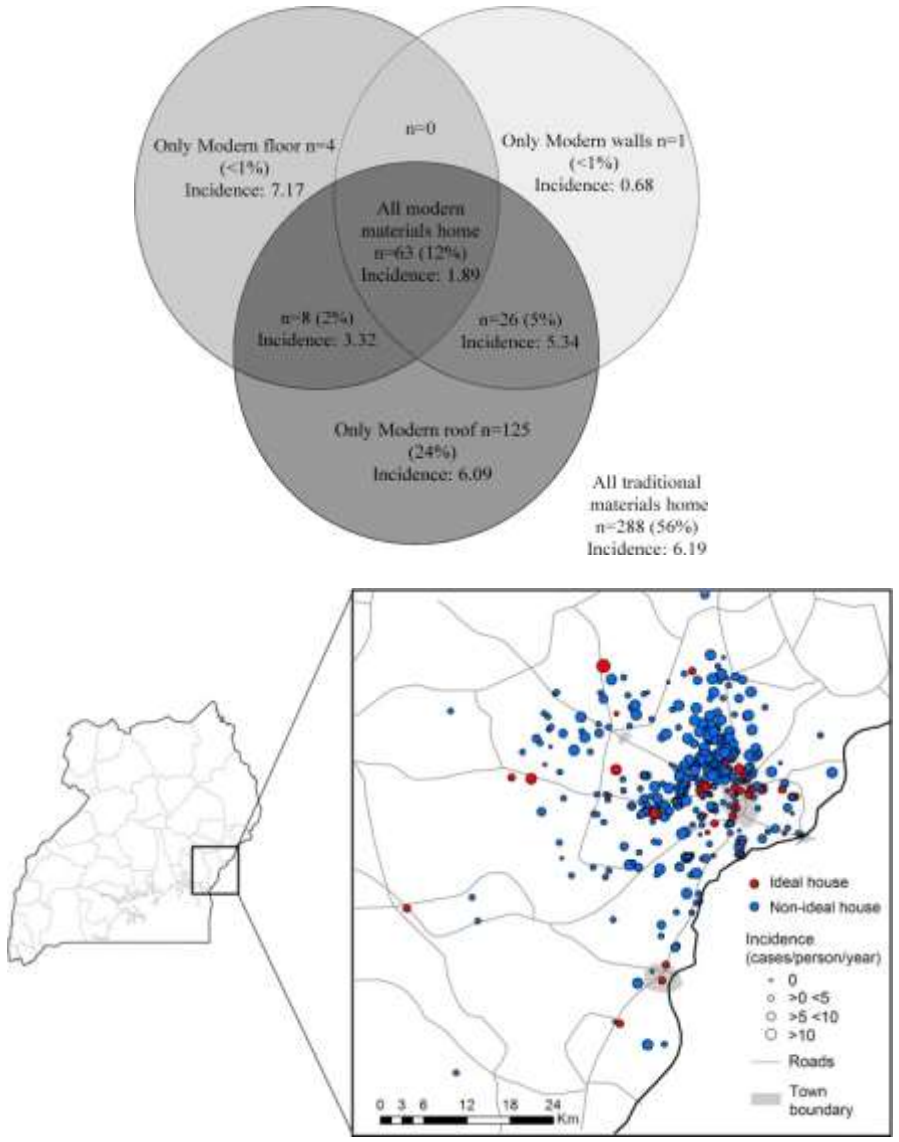
https://apps.who.int/iris/bitstream/handle/10665/259002/WHO-HTM-GVCR-2017.01-eng.pdf

What is the role of ecological and entomological surveillance in understanding urban malaria transmission?

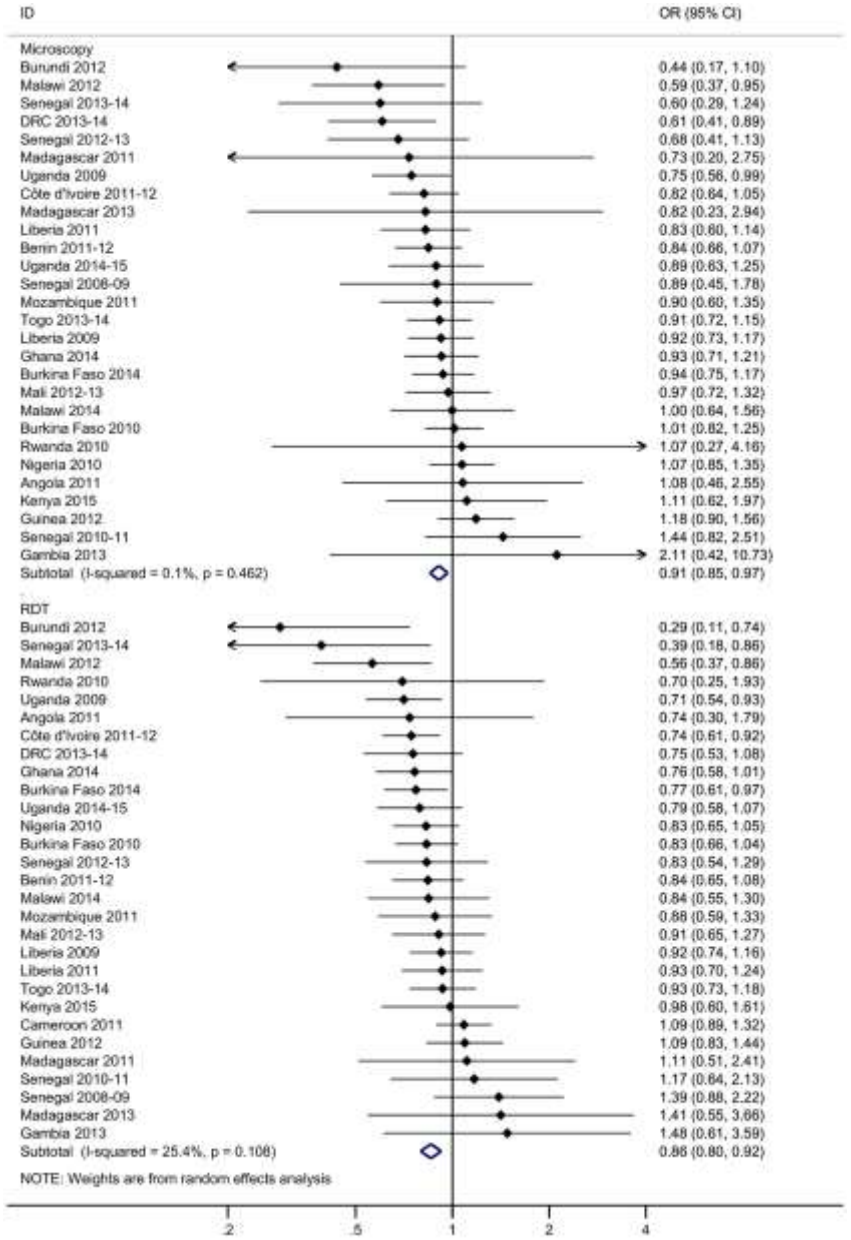
	Intervention	Has WHO malaria policy recommendation?
Environmental modification	• Improving drainage	No
	• Draining swamps	No
	• Dredging to increase water flow	No
	• Making embankments	No
	• Land reclamation	No
	• Deforestation/afforestation	No
	• Flood control	No
	• Improved sanitation including better water storage and provision and good maintenance of piped water	No No No
	• General infrastructure development – e.g. construction of roads	No No
Social/ preventive	• House/window screening	No
	• Improved housing	No
	• House inspections to identify and remove breeding sites	No No
Vector control	• Larviciding	Yes
	• Aerial spraying of insecticides	No
	• Indoor residual spraying with insecticides	Yes
	• Insecticide-treated bed nets	Yes
	• Entomological surveillance of breeding sites	Yes
	• Monitoring of insecticide resistance	Yes
Antimalarial drugs	• Early diagnosis and treatment by health care providers	Yes
	• Prophylaxis	No
	• Intermittent prevention in children	Yes (in some settings)
	• Treatment at home/ informal sector to increase access	No
	• Mass screen and treat	

What are the current approaches for malaria prevention and treatment in urban settings and which ones have a WHO policy recommendation?

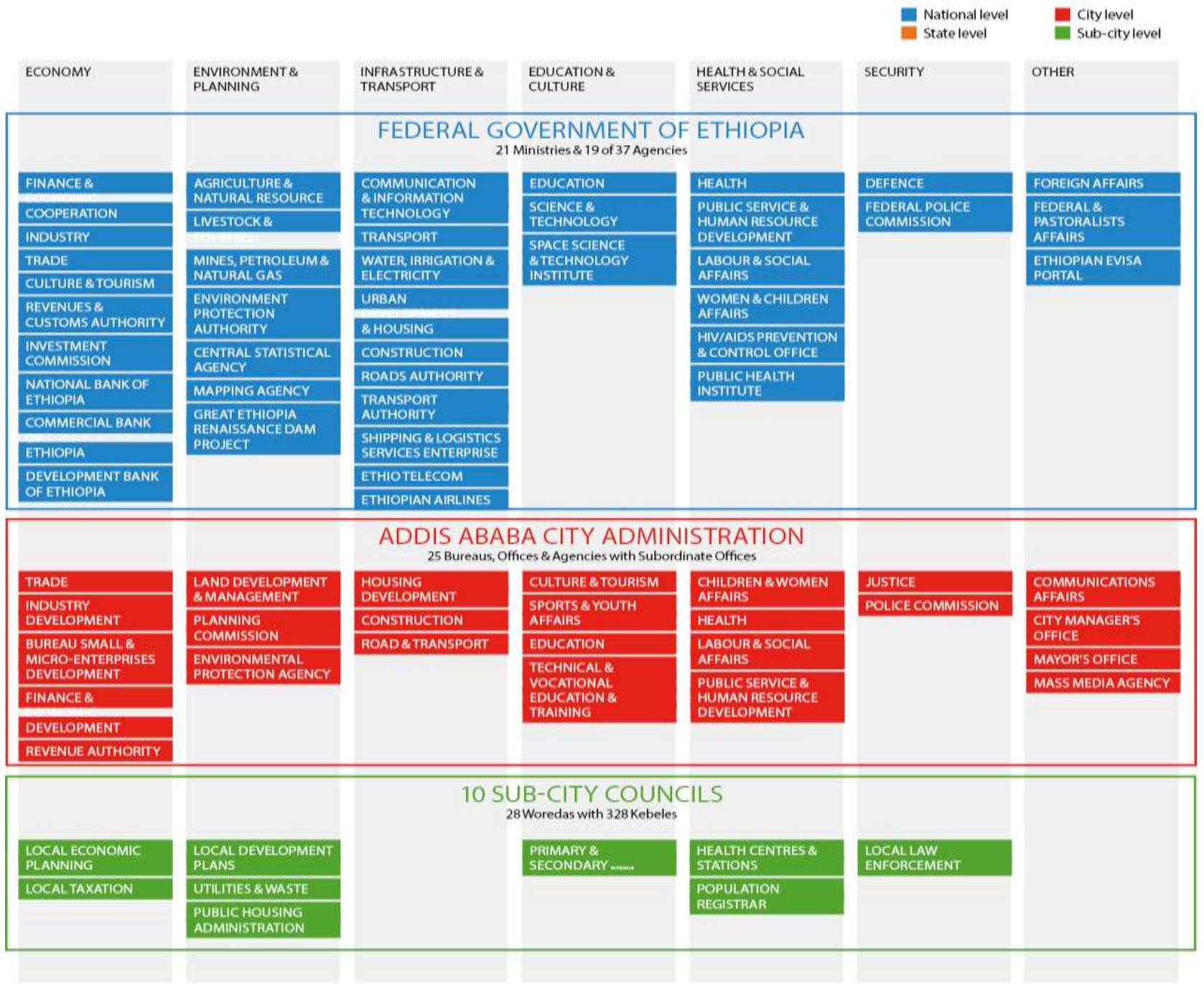
Tororo district, Uganda



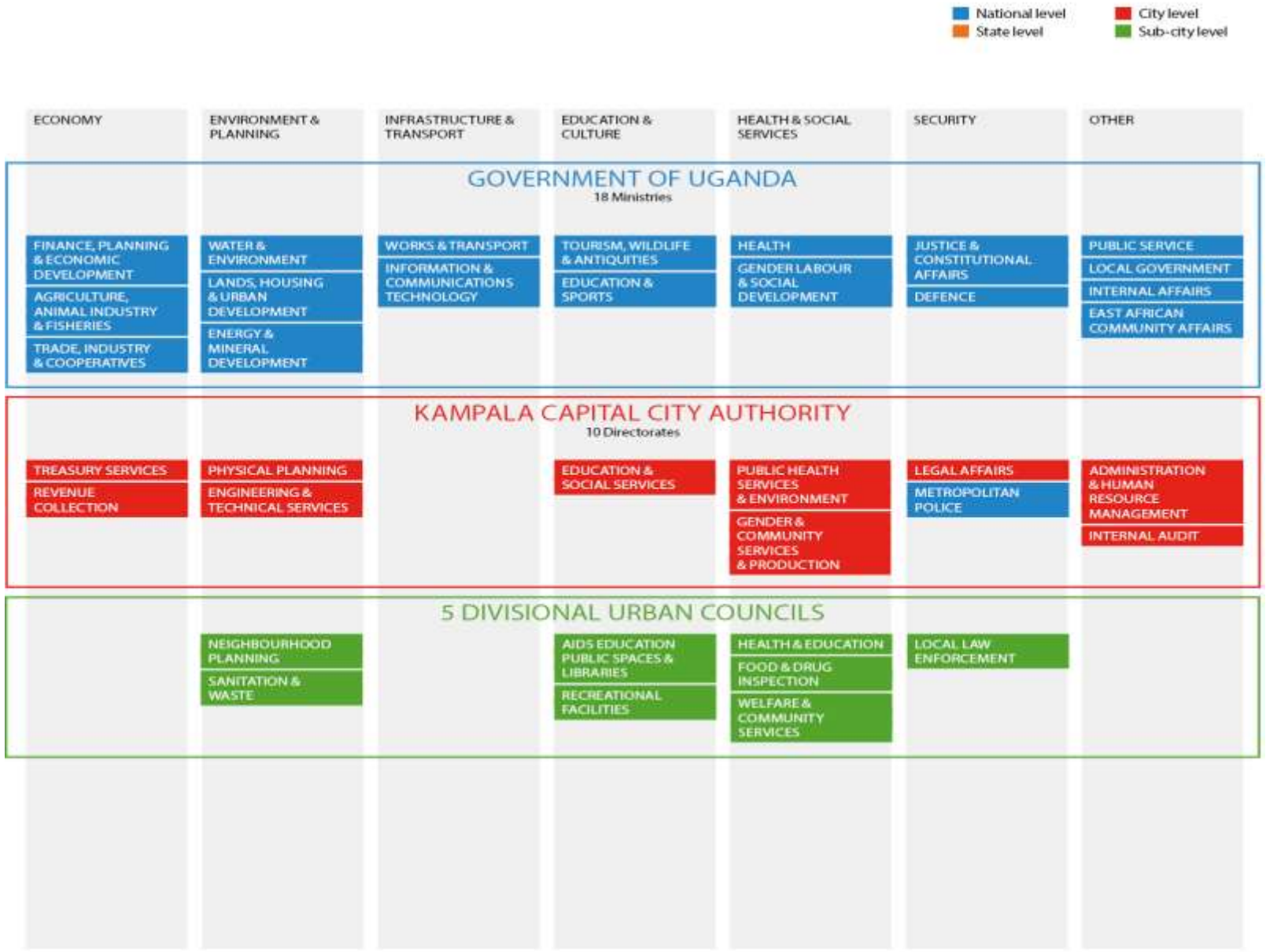
Synman *et al* (2015). Poor Housing Construction Associated with Increased Malaria Incidence in a Cohort of Young Ugandan Children, *AJTMH*



Tusting et al (2017). Housing Improvements and Malaria Risk in Sub-Saharan Africa: A Multi-Country Analysis of Survey Data. *PMED*



How do we ensure urban governance and leadership in malaria endemic countries prioritize malaria as part of the broader urban development and health systems?



https://urbanage.lsecities.net/data/governance-structure-kampala

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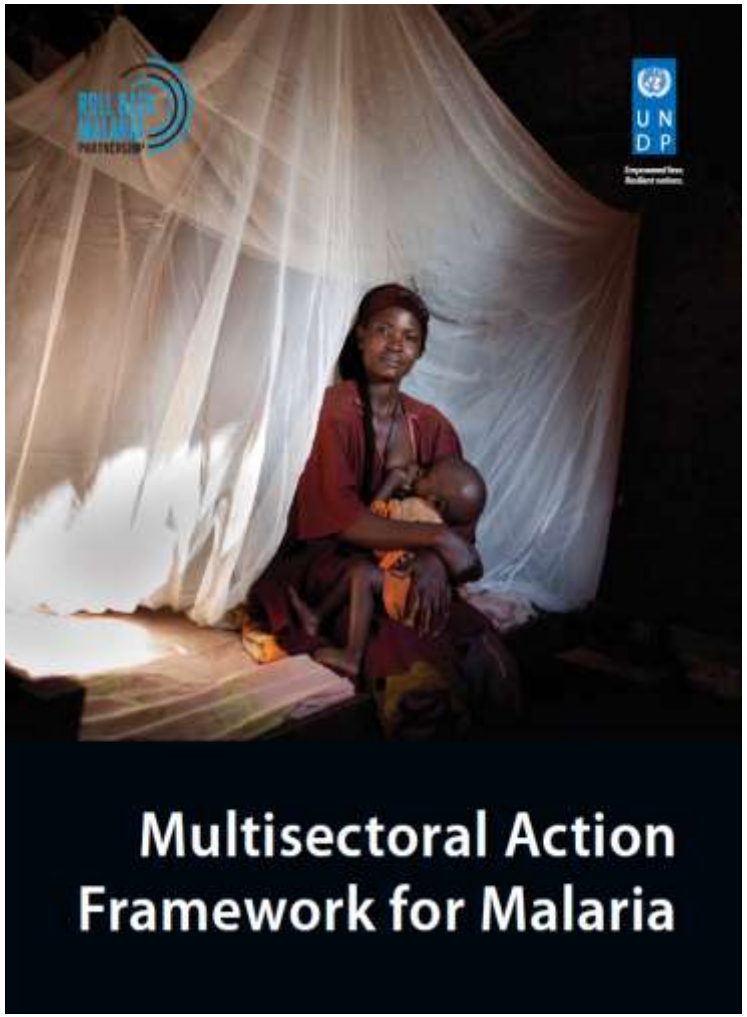
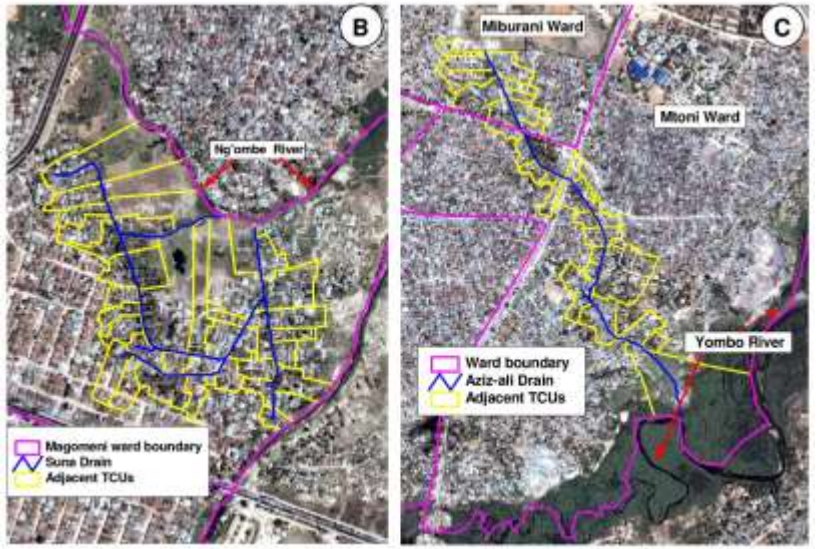


Community-based environmental management for malaria control: evidence from a small-scale intervention in Dar es Salaam, Tanzania



Castro *et al.* (2009)

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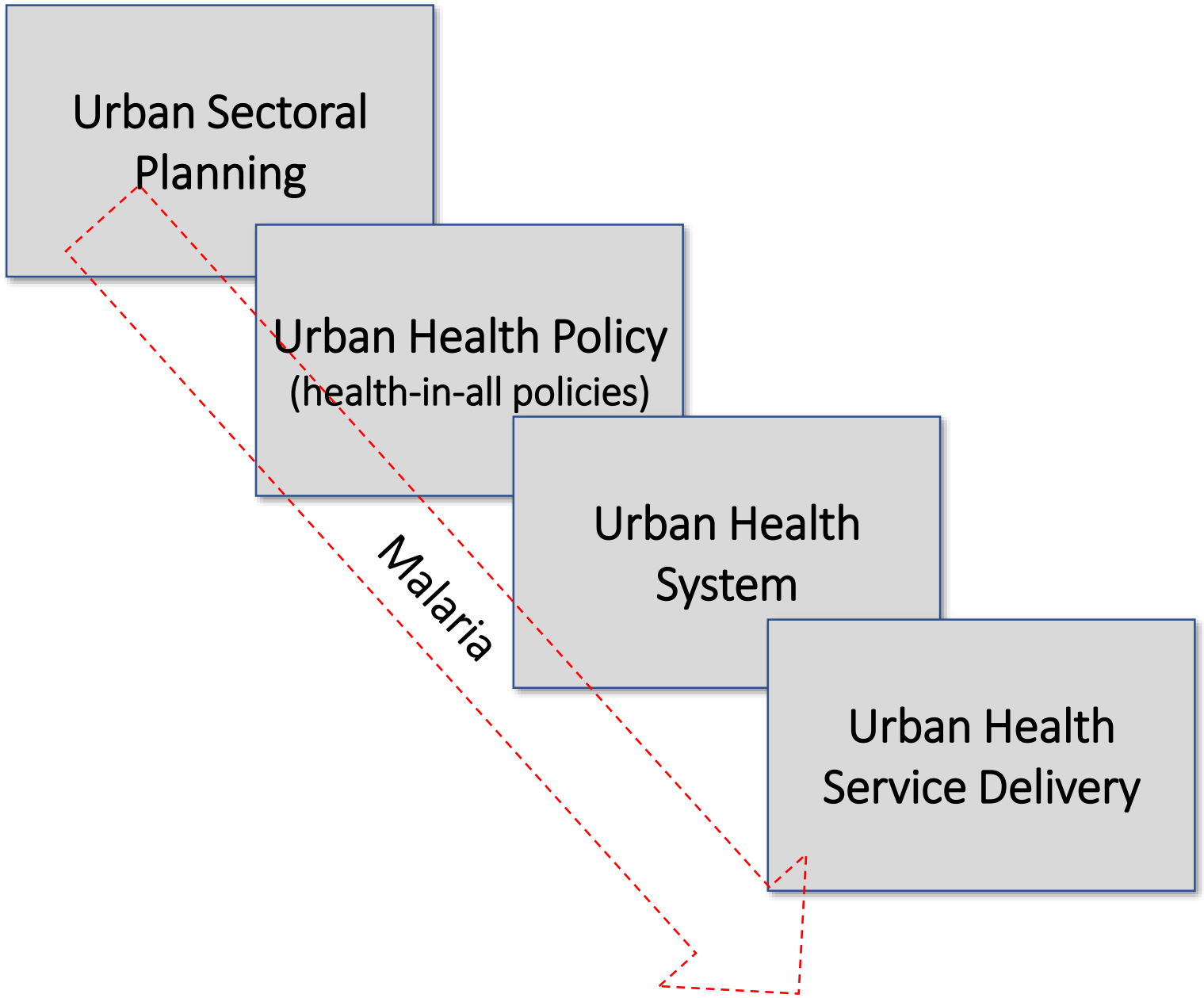

What opportunities do urban planning, health policies and health systems provide in the malaria response?

Subprogramme 1: Urban Legislation, Land, and Governance
Subprogramme 2: Urban Planning and Design
Subprogramme 3: Urban Economy and Municipal Finance
Subprogramme 4: Urban Basic Services
Subprogramme 5: Housing and Slum Upgrading
Subprogramme 6: Risk Reduction and Rehabilitation
Subprogramme 7: Urban Research and Capacity Development

What opportunities do urban planning, health policies and health systems provide in the malaria response?

Dagenais et al. Infectious Diseases of Poverty (2018) 7:85
https://doi.org/10.1186/s40249-018-0463-y

Infectious Diseases of Poverty

RESEARCH ARTICLE Open Access

Establishing research priorities in prevention and control of vector-borne diseases in urban areas: a collaborative process

Christian Dagenais[1*], Stéphanie Degroote[2], Mariam Otmani Del Barrio[3], Clara Bermudez-Tamayo[4,5] and Valéry Ridde[2,6]

Abstract

Background: In 2015, following a call for proposals from the Special Programme for Research and Training in Tropical Diseases (TDR), six scoping reviews on the prevention and control of vector-borne diseases in urban areas were conducted. Those reviews provided a clear picture of the available knowledge and highlighted knowledge gaps, as well as needs and opportunities for future research. Based on the research findings of the scoping reviews, a concept mapping exercise was undertaken to produce a list of priority research needs to be addressed.

Methods: Members of the six research teams responsible for the "VEctor boRne DiseAses Scoping reviews"

Table 2 Statements with the highest "Priority" and "Policy relevance" ratings

No.	Statements	Cluster	Priority	Policy relevance
3	The effectiveness of integrated vector control management	Interventions	4.28	4.56
1	What determines the success, effectiveness, and sustainability of preventive strategies	Interventions	4.06	4.22
26	What surveillance systems are needed to predict the next outbreaks of VBDs	Surveillance	4.06	4.00
18	How to apply the social determinant approach in integrated vector management	Equity	3.94	3.94
22	What are the impacts of interventions on health outcomes at the community level	Interventions	3.94	3.94
57	What are the ethical dimensions we need to take into account in interventions	Ethics	3.89	4.00
84	How to take into account equity in surveillance and in interventions	Equity	3.89	4.00
13	What are the sanitation waste management strategies that can help prevent VBDs	City responsibility	3.83	4.22
54	How to take social acceptability into account when designing an intervention	Community & Society	3.83	4.28
79	Barriers and facilitators for environmental sustainability of integrated vector management	Interventions	3.78	4.17

VBDs Vector-borne diseases

What are the key knowledge gaps, the priority research questions in malaria transmission dynamics and response?

TDR VEctor boRne DiseAses Scoping reviews (VERDAS) consortium

Proposed method of work

Thematic areas

Prevention interventions and delivery (vector control, chemoprevention)	Health care delivery (public, private, community)	Urban governance and policies (sectoral policies and planning, health-in-all approaches, resource allocation)	Public-private sector and community participation (resource mobilization, environmental management, social behavioural change communication etc)	Geospatial surveillance and analysis (ecology, entomology, epidemiology, microstratification and modelling)

Research priorities

- A group of 4-5 people, including at least one WHO representative and a rapporteur, will be assigned to each thematic area
- A chair will be selected by each thematic group
- Weekly 2-hour meetings will be used by thematic groups to develop key topics and content
- Each month, WHO will convene a 3-hour meeting of all thematic groups for progress updates from each group and feedback from members of the technical consultation

Timeline

- **April 2021** – call for membership
- **May 2021 (first week)** - selection
- **May 2021 (2nd or 3rd week)** – first meeting to set agenda, topics and decide on members of thematic groups
- **August 2021 (end)** – completion of meetings and summary reports from thematic areas
- **September 2021 (end)** – draft urban malaria response document
- **November 2021 (end)** – final document