Retrospective and cross-sectional study on the prevalence of malaria in selected areas under elimination program in Dembiya district, North-western Ethiopia

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Research

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Abstract

Background: Ethiopia embarked on combating malaria with an aim to eliminate malaria from low transmission districts by 2030. This involves malaria vector interventions by implementing mainly indoor residual spraying (IRS) and long-lasting insecticidal nets (LLINs) against endophilic and endophagic female Anopheles mosquitoes. Limited published reports are available about the status of malaria in areas under malaria elimination program in Ethiopia. This study intended to assess the prevalence of malaria in selected areas with a long history of implementing malaria prevention and elimination strategies.

Methods: A cross-sectional parasitological survey was conducted in two selected malaria endemic areas in Dembiya District, Northwest Ethiopia. Thin and thick blood smears collected from 735 randomly selected individuals were microscopically examined for malaria parasites. Six years retrospective malaria data was also collected from the medical records of the health centres. Structured questionnaires were prepared to collect information about the socio-economic data of the population. The data were analysed using SPSS version 20 and p < 0.05 were considered statistically significant.

Results: The six-year retrospective malaria prevalence trend indicates an overall malaria prevalence of 22.4%, out of which Plasmodium falciparum was the dominant species. From a total of 735 slides examined for the presence of malaria parasites, 3.5% (n=26) were positive for malaria parasites, in which P. falciparum was more prevalent (n=17; 2.3%), P. vivax (n=5; 0.7%), and mixed infections (n=4; 0.5%). Males were 2.6 times more likely to be infected with malaria than females (AOR = 2.6; 95% CI: 1.0, 6.4), and individuals with frequent outdoor activity were 16.4 times more vulnerable than individuals with limited outdoor activities (AOR= 16.4, 95% CI: 1.8, 147.9). Furthermore, awareness about malaria transmission was significantly associated with the prevalence of malaria.

Conclusions: Malaria is still a public health problem in Dembiya district irrespective of the past and existing vector control interventions. A malaria elimination plan might not be successful unless other alternative intervention tools targeting outdoor malaria transmission are included. For this, continuous monitoring of vectors’ susceptibility, density, and behaviour is very important in such areas.

Background

Malaria is a leading public health problem in Ethiopia, which accounts for 12% of outpatient consultations and 10% of health facility admissions [1]. More than half of the population in the country (60%) lives in malarious areas, and an estimated 68% of the total population is at risk of malaria infection [2, 3]. The transmission of malaria in Ethiopia is seasonal and unstable, and it varies with altitude and rainfall. In most parts of the country, peak malaria transmission occurs after the main rainy season (July to September). In addition, many areas experience a second minor malaria transmission period following a short rainy season from February to March [1, 2]. Most of the malaria transmissions in Ethiopia occurs in areas below 2000 m.a.s.l, but endemic regions greater than 2000 m are also documented [4, 5].

Plasmodium falciparum and P. vivax are the dominant malaria parasite species in Ethiopia, which are responsible for 60% and 40% of malaria cases, respectively [6–8]. However, P. vivax may be more dominant in different localities of the country with cooler climates [9]. In Ethiopia, Anopheles arabiensis is the primary vector of malaria, whereas An. pharoensis, An. funestus and An. nili are secondary vectors in different parts of the country [10].

The government of Ethiopia has made a massive scale-up of malaria control interventions in recent years including diagnostic testing, rapid case treatment using artemisinin-based combination therapy (ACT), prevention and control of malaria among pregnant women using intermittent preventive therapy in pregnancy (IPTp), insecticide-treated bed nets (ITNs), and indoor residual spraying (IRS) [11–14]. This has led to a significant reduction of malaria mortality and morbidity in the country [12, 15]. However, the progress towards malaria elimination is hampered because of widespread drug resistance by the parasites, and insecticide resistance in the vectors [15]. This calls for repeated malaria prevalence studies in such areas with high vector control interventions, to design additional malaria control and prevention technologies [6, 16].

This malaria prevalence study was conducted in two localities in Dembiya district, Northwestern Ethiopia, which are malaria-endemic areas with longstanding vector control interventions [17]. Over the years, malaria treatment and control measures have resulted in a significant reduction of malaria in the district [18]. However, grassroots reports on the prevalence of malaria in such localities with prolonged malaria interventions are scarce. Therefore, this study evaluated the retrospective and present trend of malaria transmission in the selected localities, and identifies socio-economic factors for malaria transmission in the study area.

Methods

Study area description
This study was conducted in Dembiya District found in the North Gondar administrative zone of Amhara regional state. The district is located at 12°39’ N and 37°09’ E. Kola Diba is the capital city of the district, located 750 km north of Addis Ababa and 35 km southwest of Gondar city. The southern part of the district is bordered by Lake Tana. The district has 45 localities (the lowest administrative unit in Ethiopia). The population of Dembiya District was estimated to be approximately 271,000 in 2007, of which 50.9% (138,000) were male and 49.1% (133,000) were females. The majority of the population (91%) lives in rural areas, with most engaging in farming activities; the remaining 9% live in urban areas. The district has 49,528 rural households with 4.3 mean household sizes [19].

The elevation of Dembiya District is between 1500 to 2600 m above sea-level. The agro-ecology of the District is midland (woyna-dega) with respective mean annual minimum and maximum temperature of 11°C and 32°C and the mean annual rainfall ranges from 995 to 1175 mm. Information obtained from the district agricultural bureau indicated that the respective proportion of areas considered as plain, mountainous, valleys, and wetland is 87%, 5%, 4.8%, and 3.2%. Out of the total area of the District, 31% is cultivated land, 16% is none cultivable land, 5.6% forest and bush, 12.8% grazing, 8.1% is covered with water, 20.2% swamp and 4.3% is residential areas. The district receives bimodal rainfall, with the short rainy season from March to May and the main rainy season from June to September.

The major crops grown in the District includes teff (*Eragrostis teff*), maize (*Zea mays*), barley (*Hordeum vulgare*), red highland sorghum (*Sorghum bicolor*), and finger millet (*Eleusine Coracana*). Besides, legumes and pulses such as chickpeas (*Cicer arietinum*) and cowpeas (*Vigna unguiculata*) are also grown in the district. They also grow some cash crops like red paper, niger seed (*Guizotia abyssinica*), fenugreek (*Trigonella foenum-graecum*), black cumin (*Nigella sativa*), White cumin (*Cuminum cyminum*), black cumin (*Nigella sativa*), White cumin (*Cuminum cyminum*), and rice (*Oryza sativa*) with a limited number of farmlands.

Guramba Bata (12°21’ N and 37°20’ E, altitude < 2000m.a.s.l.) located 7 km away from Kola Diba town. A seasonal river persisted until the end of December serving as one of tributaries to Lake Tana. Guramba Bata has one health post and one health centre, 1113 households with 6008 inhabitants (2974 are male and 3034 are females) in 2017/18 G.C (unpublished health office report) (Figure 1).

Arebiya (12°20’N and 37°22’ E, altitude < 2000m.a.s.l.) is located 17 km away from Kola Diba town. The Megech River is one of the most important rivers serving as a water source during a dry season and drains into Lake Tana. Within 1976 households, the locality has a total of 8632 inhabitants (4298 are male and 4384 are females) in 2018/19G.C. There is one health post in the locality (unpublished health office report) (Figure 1).

**Study Design.** A retrospective study was conducted to determine the six-year (2012 to 2017) of malaria prevalence by reviewing reports at Guramba Bata health centre and Arebiya health post. A cross-sectional parasitological survey was conducted in the two study localities of Dembiya district (Guramba Bata and Arebiya) following the end of the long rainy season (September to October, 2018). The two study sites were selected based on their long history of implementing vector control strategies such as IRS and LLINs.

**Retrospective malaria data collection**

To assess the retrospective trend of malaria prevalence in the study areas implementing IRS and LLINs vector intervention measures, a six-year malaria retrospective data (2012-2017) was obtained from Guramba Bata health centre and Arebiya health post. A cross-sectional parasitological survey was conducted in the two study localities of Dembiya district (Guramba Bata and Arebiya) following the end of the long rainy season (September to October, 2018). The two study sites were selected based on their long history of implementing vector control strategies such as IRS and LLINs.

**Sample size determination for active case detection**

The sample size \( n \) for estimating a population proportion of a small finite population was used to determine the sample size [20].

\[
\frac{n}{N} = \frac{n_0}{N} + \frac{1}{N} \left( \frac{n_0}{N} + \frac{1}{N} \right)
\]

Where \( n \) is the minimum sample size for a small population and \( n_0 \) is the sample size for a larger population, \( N \) is the population size (\( N \) for Guramba Bata= 6008 and \( N \) for Arebiya= 8632) and \( n_0 \) is calculated using a single point proportion formula. i.e.

\[
\frac{n_0}{N} = \frac{z^2 \cdot p(1-p)}{d^2}
\]

Where, \( p = \) prevalence of malaria (50%), \( d = \) margin of error (0.05); \( Z = \) standard score corresponds to 1.96
Where $n_1$ is the sample size for Guramba Bata study site and $n_2$ is the sample size for Arebiya study site.

**Blood sample collection and prevalence study**

Blood samples were taken from 365 individuals from Guramba Bata and 370 individuals from Arebiya study sites. These individuals were randomly selected from 160 households, considering 4.3 average persons to household of the Amhara region [19].

Thick and thin smears from finger-prick blood samples were prepared from a total of 735 individuals by well-trained laboratory technicians, from randomly selected households at the end of the rainy season (October - December, 2018). All thin smears were air dried and fixed with methanol in the field. Both thick and thin blood smears were stained with 3% Giemsa solution for 30 minutes in staining jars in the laboratory. The stained slides were rinsed with tap water and placed in an upright position to dry. The stained thick and thin films were examined with 100x oil immersion objective under a light microscope. The thick blood smear samples were first examined for the presence of *Plasmodium* parasites to determine whether the sample is positive or negative. When samples were positive, thin blood smears were examined for species identification [21].

**Socio-economic survey**

A structured questionnaire was prepared to collect information about socio-economic data of the study participants while taking blood samples. Questionnaires were filled by field assistants in consultation with the head of a household during blood sample collection.

**Statistical analysis**

The data on retrospective and prospective prevalence of malaria parasites in the two study sites, different age groups, sexes, years and species type were entered using Microsoft excel data sheet and were analysed using SPSS version 20 (Armonk, NY: IBM Corp). Chi square test was used to compare the difference in frequency of malaria prevalence between independent variables (sex, localities, and age). Association between independent variables with dependent variables was analysed using bivariate logistic regression analysis. Multivariate logistic regression was used to analyse the relative contribution of each independent variable to the dependent variable at $p \leq 0.05$.

**Results**

**Retrospective trends of malaria prevalence**

Out of 2,157 individuals who visited the two health facilities seeking treatment and suspected to have malaria, 22.4% ($n= 484$) were positive for malaria parasites (Table 1). Microscopic and RDT results indicated that 19.4% ($n= 281$) individuals in Arebiya and 28.7% ($n= 203$) individuals in Guramba Bata were infected with malaria parasites during the six-year period (2012-2017).

There were significant differences in malaria cases among the age groups in both health facilities ($\chi^2=111.8$, df $=3$, $p=0.000$; $\chi^2=231.7$, df $=3$, $p=0.000$). Malaria was more prevalent in individuals between the 18-64 age groups in both health facilities. Malaria parasites were detected in 28.5% ($n= 226$) individuals in Arebiya health post, and 67.1% ($n= 143$) individuals in Guramba Bata health centre in the 18-64 age group (Table 1). On the other hand, relatively low number of malaria cases was recorded in the 6-17 years age groups (7.2% in Arebiya health post and 9.3% in Guramba Bata health centre) (Table 1). The difference in malaria cases between sexes were statistically significant in both Arebiya health post ($\chi^2= 102.3$, df $=1$, $p=0.000$) and Guramba Bata study sites health centre ($\chi^2= 21.7$, df $=1$, $p= 0.000$). Higher malaria cases were recorded in males (27.3% and 35.9%, respectively) than in females during the six-year period in both health facilities (Table 1). Furthermore, *P. falciparum* was detected in individuals of all age groups, but it was predominant in individuals between the 18-64 years age group (23.3% and
53.3% in Arebiya and Guramba Bata respectively). *P. vivax* was frequently recorded in children less than 5 years of age group in both study localities (Table 1).

The lowest malaria cases in Arebiya health post (10.9%; n= 31) were recorded in 2016, while the highest malaria cases (41.9%; n= 72) were encountered in 2017. Similarly, lower malaria cases (11.6%; n= 11) were detected in 2016 with highest (47.4%; n= 72) malaria cases were recorded in 2017 in Guramba Bata health centre (Figure 2).

*Plasmodium falciparum* was the predominant species in the study sites during the six year period (2012-2017) (Figure 3) with the highest *P. falciparum* malaria cases (35.8%; n= 116) recorded in 2017. *P. vivax* and mixed infections were recorded in relatively lower magnitude in both sites during the six-year period (Figure 3).

**Prevalence of malaria parasites from blood sample examination**

Out of the total 735 thick and thin blood smears taken from individuals who participated in the study, 3.5% (n = 26) were positive for malaria parasites. The results from the cross sectional survey indicate that there were no statistically significant difference in percent malaria prevalence between the two localities ($\chi^2 = 0.06$, df =1, p=0.814). The prevalence of malaria infection in Guramba Bata and Arebiya study areas were 3.8% (n= 14) and 3.2% (n= 12) respectively (Table 2). *Plasmodium falciparum* was the predominant malaria parasite (2.3%, n= 17) in the study area, followed by *P. vivax* (0.7%, n=5), and mixed infections (0.5%, n= 4).

The frequency of malaria infection among the age groups was statistically significant in Arebiya study site ($\chi^2 = 8.3$, df =3, p=0.040) (Table 2). Malaria was more prevalent in the age group> 15 years old at this study site (5.8%). Whereas, in Guramba Bata study site the age groups > 15 were more infected with malaria (5%) than the other, but it was not statistically significant ($\chi^2=2.32$, df =3, p= 0.509) (Table 2). Males were more infected with malaria in Arebiya (5%) and the difference in malaria case between sexes were statistically significant ($\chi^2= 4.3$, df =1, p= 0.039) (Table 2). Similarly, in Guramba Bata study sites males were more infected with malaria (4.4%) than females, though the difference was not statistically significant ($\chi^2 = 0.31$, df = 1, p= 0.579) (Table 2)

**Socio-demographic data and Malaria risk factor analysis**

Blood samples for microscopic examination were collected from 735 randomly selected individuals from the two study localities of which 50.3% (n= 370) were from Arebiya and 49.7% (n= 365) were from Guramba Bata. Males comprised 52% (n= 382) while females were 48% (n= 353) of individuals in the sample (Table 3). The age groups, below 15, 5-9, 10-14, and above 15 accounted for 7.3% (n= 54), 18.9% (n= 139), 17.8% (n= 131), and 55.9% (n= 411) of the study participants respectively. The majority of the study participants were farmers (86.7%; n= 637) and the rest (13.3%; n= 98) were merchants. Most of the study participants (45.9 %; n= 341) were not educated. All study participants were from rural areas (Table 3).

Bivariate and multivariate analysis indicated that risk factors such as sex, age, outdoor activity in the evening, awareness about malaria transmission, the frequency of LLIN distribution, and application of IRS were significantly associated with malaria prevalence (P<0.05). However, respondent's occupation, educational level, the last time respondents received IRS were not significantly associated with malaria transmission (P>0.05; Table 4).

Males were 2.6 times more likely to be infected with malaria than females (AOR = 2.6, 95% CI: 1.04, 6.41) and individuals with high outdoor activity were 16.4 times more vulnerable than individuals with limited outdoor activities (AOR= 16.4, 95% CI: 1.82, 147.85). Respondents who are not aware of malaria transmission and control were highly infected with malaria than those who were aware of it (AOR=0.3, 95% CI: 0.12-0.82). The last time respondents received LLINs (before a year) was associated with a low level of malaria prevalence in the study area (Table 4).

**Discussion**

This study evaluated the six-year retrospective data of malaria prevalence from health facility records in Guramba Bata and Arebiya localities, where vector control strategies such as IRS and ITN have been implemented for more than a decade. A snapshot cross-sectional malaria survey was also conducted to determine the level of malaria transmission and the malaria parasites that prevail in the study area. Results from both retrospective and cross-sectional studies indicate that malaria is a public health problem in the area. The overall percent malaria cases detected in the retrospective malaria study were 22.4% (n = 484) with percent malaria cases peaking towards 2017 (44.4%) despite of the ongoing IRS and LLINs malaria vector control strategies implemented in the study area. A relatively higher percent of malaria cases were reported from a similar retrospective study in the nearby Kola Diba health centre (39.6%) [22], and Serbo health centre (43.8%) [23]. However, this result is higher than a retrospective study conducted in Metema hospital [24] and Kombolcha [25], where the prevalence was 17%, and 7.5% respectively.
The overall prevalence of malaria from this cross-sectional study was 3.5% (26/735) and \textit{P. falciparum} was the predominant malaria parasite, this is comparable with the 3.9% prevalence reported from a cross-sectional study conducted in Awassa Town [26]. The result of the cross-sectional study was lower than the 5.3% prevalence of malaria reported in Gondar Town [27], and the 5.2% malaria prevalence from Jimma Town [28].

The six-year retrospective data indicated that malaria prevalence varied from year to year, with relatively lower malaria cases recorded in 2013, 2015 and 2016. The reduced number of malaria cases during these years could be associated with the accumulated effect of scaled-up malaria control interventions and improved community awareness in the study area. The relatively higher number of malaria cases reported in 2017 indicated an increase in malaria prevalence in the study area, irrespective of continued malaria vector control interventions. This is in agreement with the existing scenarios advocating that malaria remains a public health problem in Ethiopia even though intensive vector intervention strategies were implemented [29]. This could possibly be because of inappropriate utilization of vector intervention strategies, the development of insecticide resistant malaria vector and development of resistance in \textit{Plasmodium} parasites to the existing drugs.

The species specific prevalence in this study showed that \textit{P. falciparum} was the dominant, whereas \textit{P. vivax} and mixed prevalence holds the second and third position respectively. This is in line with the fact that \textit{P. falciparum} is the dominant parasite in many parts of Ethiopia with altitude below 2000 m a.s.l. [30]. Similar trend of \textit{Plasmodium} parasite distributions were reported from Gilgel-Gibe [31] and children from Northern Ethiopia [32] and migrant laborers from North-western Ethiopia [33]. Retrospective studies from Kola Diba health centre [22], Serbo health centre [23], Metema hospital [24], Kombolcha [25], and Tselemti Woreda [34] and a survey from different part of Ethiopia [35] also support this study finding. However, reports showed that \textit{P. vivax} was the dominant species in some part of Ethiopia [28, 36, & 37].

The retrospective and prospective studies indicated that malaria infections were more prevalent in males than in females. Similar studies indicated that males were more infected with malaria than females in different part of Ethiopia [38–41] and in Kenya [42]. It was presumed that individual behaviour, environmental and socio-economic factors contribute to transmission of malaria in Ethiopia [43, 44]. Likewise, malaria was more prevalent in individuals above the age group of \( \geq 15 \). This is in agreement with a retrospective study conducted in Kombolcha [25] and Kola Diba health centres [22]. Male individuals at these productive ages are actively involved in outdoor activities such as agriculture and cattle herding, in the evening which makes them vulnerable to outdoor \textit{Anopheles} mosquito biting. Male individuals usually spend the night outside the house tending cattle in the study sites. These outdoor activities at night were predictors associated with malaria transmission in the study areas. Similarly, different reports indicated that outdoor activities in the evening contributed to high malaria transmission [41, 43] mainly due to the fact that individuals with outdoor activities are exposed to outdoor biting by \textit{Anopheles} mosquitoes [45]. During this study, a substantial number of children were also infected with \textit{Plasmodium} parasites during this study, which indicates the endemicity of malaria in the study area.

The current study showed that poor awareness and knowledge about malaria prevention and control contributed to the prevailing malaria transmission in the study area. Similar reports indicated that awareness about malaria was associated with malaria transmission in Ethiopia [41] and Kenya [42]. This urges for a continuous need to educate and increase awareness of the local communities about malaria transmission towards improved malaria prevention and control strategies.

**Conclusions**

The prevalence of malaria in Dembiya District, where vector control strategies such as long-lasting insecticide treated bed nets and indoor residual sprays have been implemented for over three decades, was determined using prospective and retrospective data. The respective overall prevalence of \textit{Plasmodium} infection from a retrospective and prospective study was 22.4% and 3.5%, indicating malaria remains a public health problem in the district. Factors such as outdoor activity and poor knowledge about malaria transmission contributed to malaria infection in the district. Therefore, the national malaria elimination program would not be possible unless other additional alternative of malaria prevention and control methods targeting outdoor vector control and awareness creation are designed. In addition, a continuous monitoring of vector ecology and behaviour is important to design a holistic approach to malaria control strategies.

**Abbreviations**

LLINs: Long lasting insecticide treated bed nets

IRS: Indoor residual spray

CI: Confidence interval

\( \chi^2 \): chi square
df: Degree of freedom

RDT: Rapid diagnosis test

IPTp: intermittent preventive therapy in pregnancy

Declarations

Ethics approval and consent to participate

Ethical clearance was obtained from Addis Ababa University, institutional ethical review board of the College of Natural and Computational Sciences (Ref No CNSDO/692/10/2018). Written consent was obtained from the head of the household and other study participants before sampling. Individuals proved to be positive for malaria during blood film examination were treated with antimalarial drug prescribed by physicians.

Consent for publication

Not applicable

Availability of data and materials

The data sets supporting the conclusions of this article are available at Addis Ababa University College of Natural and Computational Sciences.

Competing Interest

The authors declare that there is no conflict of interest.

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Authors’ contributions

MT, HT, YW and SD designed the study. HT, YW and SD supervised and MT conducted the experiments. MT conducted the statistical analyses. MT developed first draft, HT, YW and SD revised the manuscript. All authors read and approved the final manuscript.

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Tables

Table 1. A six years retrospective trend of malaria cases in the two localities of Dembiya District (2012- 2017).
### Table 2. Prevalence of malaria from the cross-sectional study in the two localities of Dembiya District (October-November, 2018).

| Study sites | Arebiya | Guramba Bata |
|-------------|---------|--------------|
| Age         |         |              |
| < 5         |         |              |
| No. Examined | 63      | 66           |
| P.f (%)     | 2(3.2)  | 9(13.6)      |
| P.v (%)     | 4(6.3)  | 8(12.1)      |
| Mixed (%)   | 0       | 0            |
| Total +ve (%) | 6(9.5)  | 17(25.8)     |
| 6-17        |         |              |
| No. Examined | 553     | 418          |
| P.f (%)     | 23(4.2) | 23(5.5)      |
| P.v (%)     | 17(3.1) | 8(1.9)       |
| Mixed (%)   | 0       | 8(1.9)       |
| Total +ve (%) | 40(7.2) | 39(9.3)      |
| 18-64       |         |              |
| No. Examined | 792     | 213          |
| P.f (%)     | 187(23.6) | 114(53.5)  |
| P.v (%)     | 27(3.4) | 20(9.4)      |
| Mixed (%)   | 12(1.5) | 9(4.2)       |
| Total +ve (%) | 226(28.5)| 143(67.1)   |
| >65         |         |              |
| No. Examined | 41      | 11           |
| P.f (%)     | 9(21.9) | 0            |
| P.v (%)     | 0       | 4(36.2)      |
| Mixed (%)   | 0       | 0            |
| Total +ve (%) | 9(21.9) | 4(36.4)      |
| Total       | 1449    | 708          |
|              | 221(15.3)| 146(20.6)   |
|              | 48(3.3) | 40(5.6)     |
|              | 12(0.8) | 17(2.5)     |
|              | 281(19.4)| 203(28.7)   |
| Sex         |         |              |
| Male        |         |              |
| No. Examined | 931     | 398          |
| P.f (%)     | 207(22.2)| 110(27.6)   |
| P.v (%)     | 36(3.9) | 23(5.8)     |
| Mixed (%)   | 11(1.2) | 10(2.5)     |
| Total +ve (%) | 254(27.3)| 143(35.9)   |
| Female      |         |              |
| No. Examined | 518     | 310          |
| P.f (%)     | 14(2.7) | 36(11.6)    |
| P.v (%)     | 12(2.3) | 17(5.5)     |
| Mixed (%)   | 1(0.2)  | 7(2.3)      |
| Total +ve (%) | 27(5.2) | 60(19.4)    |
| Total       | 1,449   | 708          |
|              | 221(15.3)| 146(20.6)   |
|              | 48(3.3) | 40(5.6)     |
|              | 12(0.8) | 17(2.4)     |
|              | 281(19.4)| 203(28.7)   |

*= χ²=111.8, df =3, p=0.000; ** = χ²=231.7, df =3, p=0.000; ‡ = χ²= 102.3, df =1, p=0.000; † = χ²= 21.7, df = 1, p=0.000
| Study sites | Arebiya | Guramba Bata |
|-------------|---------|-------------|
| **Age**     | **No. Examined** | **P.f (%)** | **P.v (%)** | **Mix (%)** | **Total +ve (%)** | **Age**<br>**No. Examined** | **P.f (%)** | **P.v (%)** | **Mix (%)** | **Total +ve (%)** |
| < 5         | 30      | 0           | 0           | 0           | 0 | 0 | < 5 | 24 | 1(4.2) | 0 | 0 | 1(4.2) |
| 5-9         | 76      | 0           | 0           | 0           | 0 | 0 | 5-9 | 63 | 1(1.6) | 0 | 0 | 1(1.6) |
| 10-14       | 74      | 1(1.4)      | 0           | 0           | 1(1.4) | 10-14 | 57 | 0 | 1(1.8) | 0 | 0 | 1(1.8) |
| ≥15         | 190     | 6(3.2)      | 2(1.1)      | 3(1.6)      | 11(5.8) | ≥15 | 220 | 8(3.6) | 2(0.9) | 1(0.5) | 11(5) |
| **Total**   | 370     | 7(1.9)      | 2(0.5)      | 3(0.8)      | 12(3.2) | **Total** | 365 | 10(2.7) | 3(0.8) | 1(0.3) | 14(3.8) |
| **Sex**     | **No. Examined** | **P.f (%)** | **P.v (%)** | **Mix (%)** | **Total +ve (%)** | **Age**<br>**No. Examined** | **P.f (%)** | **P.v (%)** | **Mix (%)** | **Total +ve (%)** |
| Male        | 200     | 6 (3)       | 1           | 3 (1.5)     | 10 (5) | Male | 182 | 4 (2.2) | 3 | 1 (0.5) | 8 (4.4) |
| Female      | 170     | 1           | 1           | 0           | 2 (1.2) | Female | 183 | 6 (3.3) | 0 | 0 | 6 (3.3) |
| **Total**   | 370     | 7           | 2           | 3 (0.8)     | 12 (3.2) | **Total** | 365 | 10 | 3 | 1 (0.3) | 14 (3.8) |

\[* = \chi^2 = 8.3, df = 3, p=0.040; ** = \chi^2=2.32, df =3, p= 0.509; \ddot{=} = \chi^2 = 4.3, df =1, p= 0.039; \dddot{=} = \chi^2= 0.31, df = 1, p= 0.579\]

**Table 3. Socio-demographic data of the study participants in the two localities of Dembiya District.**
| Variables          | Study sites | Total (%) |
|--------------------|-------------|-----------|
| Sex                | Arebiya    | Guramba Bata |
| Male               | 200        | 182       | 382 (52) |
| Female             | 170        | 183       | 353 (48) |
| Total              | 370 (50.3%) | 365 (49.7%) | 735 |
| Age                |             |           |         |
| <5                 | 30          | 24        | 54 (7.3) |
| 5-9                | 76          | 63        | 139 (18.9) |
| 10-14              | 74          | 57        | 131 (17.8) |
| ≥15                | 190         | 221       | 411 (55.9) |
| Total              | 370         | 365       | 735 |
| Occupation         |             |           |         |
| Farmer             | 332         | 305       | 637 (86.7) |
| Merchant           | 38          | 60        | 98 (13.3) |
| Total              | 370         | 365       | 735 |
| Educational status |             |           |         |
| No formal education| 146         | 195       | 341 (46.4) |
| Primary school attendees | 119 | 104 | 223 (30.3) |
| Secondary school attendees | 71 | 48 | 119 (16.2) |
| More than secondary | 34          | 18        | 52 (7.1) |
| Total              | 370         | 365       | 735 (100) |

Table 4. Bivariate and multivariate analysis of factors associated with malaria infection in selected localities of Dembiya District.
| Variables              | Category      | Total Examined | Positive (%) | OR (95% CI)          | COR            | AOR            | p-value |
|-----------------------|---------------|----------------|--------------|----------------------|----------------|----------------|---------|
|                       |               |                |              |                      |                |                |         |
| Sex                   | Female        | 353            | 8 (2.3)      | 1                    | 1              | 1              |         |
|                       | Male          | 382            | 18(4.7)      | 2.13(0.92-4.97)*     | 2.58(1.04-6.41) | 0.041          |         |
| Age                   | <5            | 54             | 0            | 1                    | 1              |                |         |
|                       | 5-9           | 139            | 1(0.7)       | 0.38(0.02-6.252)*    | 0.31 (0.02-5.27) | 0.417          |         |
|                       | 10-14         | 131            | 3(2.3)       | 0.82(0.07-9.26)      | 0.53 (0.04-6.48) | 0.617          |         |
|                       | ≥15           | 413            | 22(5.3)      | 2.99(0.39-22.69)     | 2.15(0.27-16.92) | 0.466          |         |
| Occupation            | Farmer        | 637            | 25(3.9)      | 3.96(0.53-29.58)     | 4.16(0.49-35.22) | 0.191          |         |
|                       | Merchant      | 98             | 1(1)         | 1                    | 1              |                |         |
| Educational status    | No education  | 341            | 14 (4.1)     | 1.07(0.24-4.85)      | 1.19(0.24-5.93) | 0.837          |         |
|                       | Primary       | 223            | 6 (2.7)      | 0.69(0.14-3.53)      | 0.87 (0.16-4.74) | 0.873          |         |
|                       | Secondary     | 119            | 4 (3.4)      | 0.87(0.15-4.90)      | 1.58(0.25-10.02) | 0.629          |         |
|                       | > Secondary   | 52             | 2 (3.8)      | 1                    | 1              |                |         |
| Outdoor activity      | Yes           | 571            | 25 (4.4)     | 7.46(1.00-55.50)*    | 16.42(1.82-147.85) | 0.013          |         |
|                       | No            | 164            | 1 (0.6)      | 1                    | 1              |                |         |
| Awareness about       | Yes           | 441            | 12 (2.7)     | 1                    | 1              |                |         |
| transmission          | No            | 286            | 14 (4.9)     | 1.87(0.85-4.11)*     | 3.17(1.22-8.24) | 0.018          |         |
| Period of receiving   | <6 month      | 541            | 20 (3.7)     | 1                    | 1              |                |         |
| last IRS              | 6-12 month    | 194            | 6 (3.1)      | 0.83(0.33-2.10)      | 1.98(0.66-5.91) | 0.221          |         |
|                       | More than 6   | 16             | 3 (18.8)     | 1                    | 1              |                |         |
| Period of receiving    | 6-12 month    | 15             | 4 (26.7)     | 1.58(0.288-8.61)     | 3.32(0.47-23.74) | 0.231          |         |
| last LLIN             | More than 1   | 704            | 19 (2.7)     | 0.12(0.032-0.46)*    | 0.12(0.02-0.57) | 0.008          |         |

*: Indicates significant values at p≤0.05

**Figures**
Figure 1

Study area map. Note: The designations employed and the presentation of the material on this map do not imply the expression of any opinion whatsoever on the part of Research Square concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. This map has been provided by the authors.
Figure 2

Six-year annual percent malaria cases reported in Arebiya and Guramba Bata (2012 - 2017).
Figure 3

Six-year annual species specific prevalence of malaria parasites reported in Arebiya and Guramba Bata (2012 - 2017).