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Therapeutic efficacy of artemether–lumefantrine in the treatment of uncomplicated *Plasmodium falciparum* malaria in Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia

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Abstract

Background Artemether–lumefantrine (AL) has been the primary anti-malarial drug used to treat uncomplicated *Plasmodium falciparum* malaria in Ethiopia since 2004. However, there have been recent reports of AL resistance mutations in different African countries, including Ethiopia. This is concerning and requires periodic monitoring of anti-malarial drug resistance. Therefore, the current study aimed to evaluate the therapeutic efficacy of AL in treating uncomplicated *P. falciparum* malaria in the Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia.

Methods A single-arm prospective study with a 28-day follow-up period was conducted from July to October 2022. Capillary blood samples were collected for RDT and microscopic examination. The study enrolled monoinfected *P. falciparum* patients aged ≥ 18 years at Ganta Sira Health Post. Sociodemographic and clinical data were recorded, and a dried blood spot (DBS) was prepared for each participant. Nested polymerase chain reaction (nPCR) genotyping of the *msp-1* and *msp-2* genes was only performed for recurrent cases to distinguish between recurrence and reinfection. Data entry and analysis were performed using the WHO Excel spreadsheet and SPSS version 26.

Results A total of 89 patients were enrolled, and 67 adequately completed the 28-day follow-up period. AL showed a 100% clearance rate for fever on day 2 and asexual parasites on day 3. Gametocytes were detected in 13.5% (12/89) of the participants. The gametocyte clearance rate was 58.3% (7/12) until day 7 and 100% (12/12) until day 14. Five participants developed recurrent malaria, three of whom experienced relapse and two of whom experienced reinfection. Based on the Kaplan–Meier survival analysis, the PCR-uncorrected and PCR-corrected cumulative incidence of success were 93.7% (95% CI 85.5–97.3) and 96.2% (95% CI 85.5–98.7), respectively.

Conclusion AL was efficacious in treating uncomplicated *P. falciparum* malaria in the study area. However, the detection of recurrent patients highlights the need for continuous efficacy studies in this area.

Keywords Cure rate, Parasite clearance, Recurrence, Malaria

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Background

Malaria parasites in humans are still a significant public health concern in malaria-endemic countries. An increase in malaria cases was reported globally between 2020 (245 million cases) and 2021 (247 million cases). In 2021, 95% of malaria cases were reported in the African region [1]. In Ethiopia, a 34% increase in malaria cases was observed in 2020, with 1,389,750 confirmed cases compared to 904,405 confirmed cases in 2019 [2]. *Plasmodium falciparum* is the dominant cause of malaria in most parts of Ethiopia [3].

Plasmodium falciparum has developed resistance to several anti-malarial drugs, such as chloroquine, sulfadoxine–pyrimethamine (SP) and mefloquine [4]. To control the spread of resistance, the World Health Organization (WHO) recommends the use of artemisinin-based combination therapy (ACT) as the first-line treatment for uncomplicated *P. falciparum* malaria in endemic countries [5]. However, the first case of artemisinin resistance in *P. falciparum* was reported in Cambodia in 2009, and it has since spread to other Southeast Asian countries [6]. More recently, artemisinin resistance has been reported in Rwanda and Northern Uganda, indicating the emergence of this resistance in Africa [7, 8]. This resistance has the potential to spread to neighboring countries and other parts of the world.

Anti-malarial drug-resistant strains of malaria can spread rapidly and cause epidemics with severe public health and economic consequences. To identify the emergence and spread of anti-malarial drug resistance, in vivo therapeutic efficacy studies are considered the gold standard method [9]. If the treatment failure rate exceeds 10%, it is recommended to change the national anti-malarial treatment policy [10].

Since 2004, artemether–lumefantrine (AL) has been the first-line drug for treating uncomplicated *P. falciparum* malaria in Ethiopia [11]. Several therapeutic efficacy studies in Ethiopia have reported a PCR-corrected cure rate of over 96% [12, 13]. However, it is recommended by the WHO to regularly monitor anti-malarial drug efficacy at least every two years in malaria-endemic countries [10] to detect any increase in drug resistance early. This approach helps to make rapid and evidence-based decisions on anti-malarial treatment policies. Therefore, this study aimed to assess the therapeutic efficacy of AL in the treatment of uncomplicated *P. falciparum* malaria in the Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia.

Methods

Study setting

This study was carried out in Ganta Sira Health Post, which is located in Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia (Fig. 1). Sille Village is situated 518 km from Addis Ababa, the capital of Ethiopia, and 13 km from Arba Minch, the capital of the Gamo Zone. The geographical location of the study area is between 5° 54′ 6.41′′ N and 5° 59′ 33.13′′ N latitude and between 37° 26′ 35.32′′ E and 37° 32′ 9.89′′ E longitude. The altitude of the study area ranges from 1120 to 1380 m above sea level. It is one of the malaria endemic villages in the Arba Minch Zuria District, with a hot and humid climate suitable for malaria vectors. The temperature ranges from 25 to 36 °C, and the average annual rainfall is between 900 and 1300 mm. The village has high irrigation potential, with the Sille River and Lake Chamo serving as water sources for irrigation.

The primary source of income in the village is agriculture, with banana serving as the main cash crop. The village's total human population was 3938 in 2022 (from the annual report of Gant Sira Health Post). There is only one health post in the village that provides basic public health services. The primary malaria control strategies in the village include indoor residual spraying, insecticide-treated nets, and case management using anti-malarial drugs (AL, chloroquine and primaquine). The rapid diagnostic test (RDT) is the main diagnostic tool used in health posts.

Study design, period and reporting

A single-arm prospective study was conducted to evaluate the efficacy of AL for treating uncomplicated *P. falciparum* malaria based on the revised WHO protocol [10] during the malaria transmission season from July to October 2022. The study is reported in line with WHO protocol guideline and STARTER checklist for anti-malarial therapeutic efficacy reporting [10, 14].

Source and study population

All individuals with suspected malaria who visited Ganta Sira Health Post for malaria diagnosis during the study period composed the source population. Patients aged ≥ 18 years and positive for *P. falciparum* mono-infection composed the study population.

Inclusion and exclusion criteria

To qualify for the study, patients must be permanent residents in the Health Post catchment area, at least 18 years old, have an axillary temperature ≥ 37.5 °C, or have had a fever within the previous 24 h. Additionally, patients must have a mono-infection with *P. falciparum,* with an asexual parasitaemia level between 1000–200,000 parasites/μl of blood. They must be able to swallow oral medication and be willing to comply with the study protocol.

The study had certain exclusion criteria, including pregnancy and breastfeeding, infection with a type of malaria

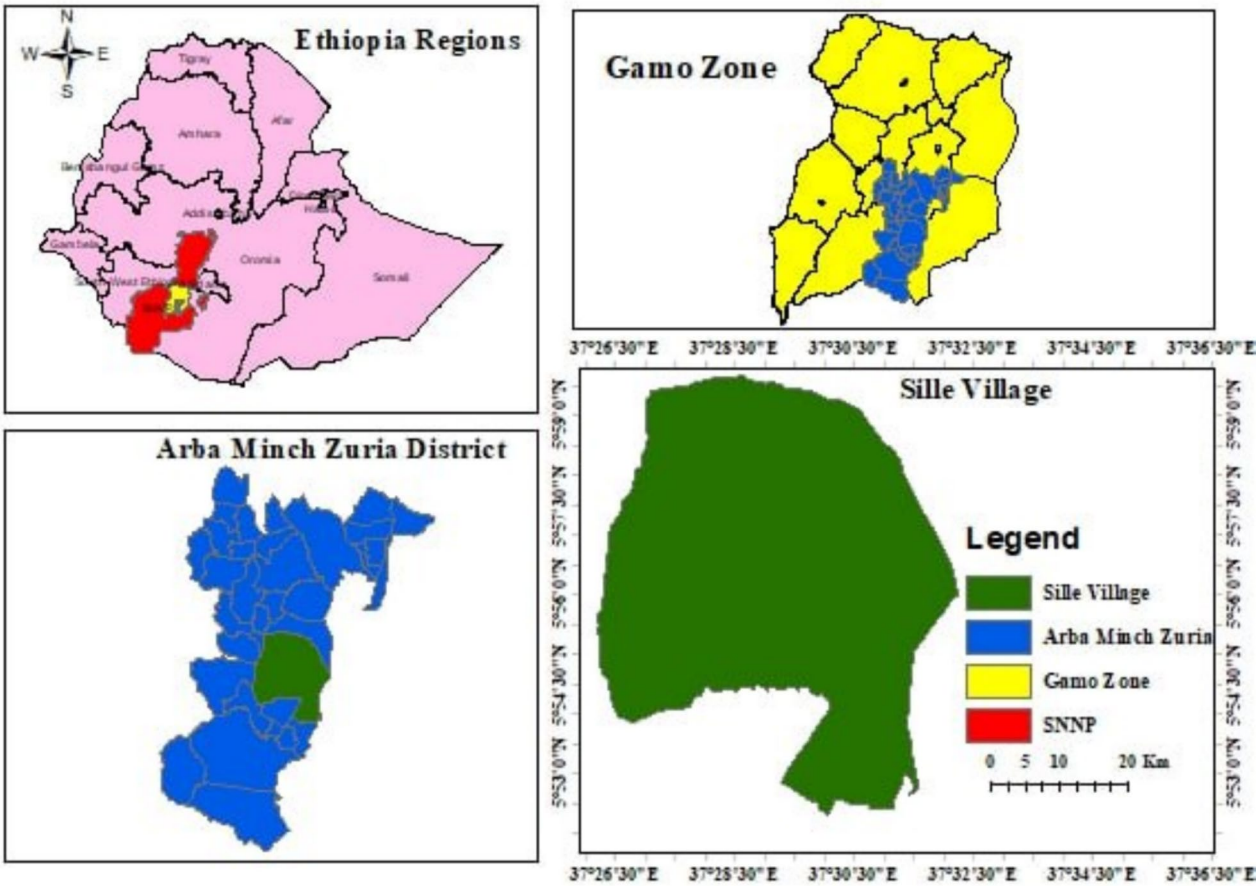

Fig. 1 Map of the study area

other than *P. falciparum*, use of an AL drug within the previous two weeks, continuous vomiting, known hypersensitivity to AL, severe malaria, and regular use of a drug that may interfere with AL pharmacokinetics.

Sample size determination

The required sample size was calculated by using a single population proportion formula based on the revised WHO protocol [10]. The sample size calculation assumed a 5% treatment failure rate for AL, with a desired precision of 5% and a confidence interval (CI) of 95%. The calculated initial sample size was 73, and assuming an additional 20% loss to follow-up, the expected sample size was 88.

Sampling technique and data collection

A consecutive sampling technique was used to select the study participants until the required sample size was reached. Sociodemographic and clinical data were recorded for each patient using structured questionnaires.

Parasitological assessment

RDT

All patients with suspected malaria who visited the Health Post were examined by a conventional RDT manufactured by Abbott Diagnostics Korea Inc., Republic of Korea, on day 0 per the manufacturer's protocol.

Blood film examination

RDT is the routine malaria diagnostic test in the health post. All patients suspected of having malaria were primarily subjected to RDT, and microscopic examination was performed for all patients by a senior laboratory technologist from South Ethiopia Region Public Health Institute, whether RDT negative or positive [10]. Discrepancies between RDT and microscopic reader were confirmed by nested PCR for which RDT negative but *P. falciparum* positive by a laboratory technologist. Thick and thin blood smears were prepared for all patients. A capillary blood sample of approximately 6 µl was used for the thick film, and 2 µl was used for the thin film. Blood smears were also obtained on each follow-up day

(1, 2, 3, 7, 14, 21 and 28 days) for the study participants. The blood smears were dried on a flat surface bench, and the thin smear was fixed with absolute methanol. Both the thick and thin blood smears were then stained with freshly prepared 10% Giemsa working solution for 10 min [15]. The samples were then examined at 1000× magnification. Thick-cell smears were reported as negative when no parasite was detected after 100 microscopic fields were examined. The parasite density at the asexual and sexual stages was determined by counting the number of parasites per 200 and 1000 white blood cells (WBCs), respectively, on thick blood films, assuming a total standard WBC count of 8000/µl [16].

The asexual and sexual stage parasite density was calculated as follows:

- $$\text{Asexual parasite density}/\mu l = \frac{\text{number of parasites counted}}{200\ \text{WBC}} \times 8000\ \text{WBC}$$
- $$\text{Gametocyte density}/\mu l = \frac{\text{number gametocytes counted}}{1000\ \text{WBC}} \times 8000\ \text{WBC}$$

Dried blood spot (DBS)

Three drops of blood on filter paper (each approximately 20 µl) were collected from the study participants on days 0, 7, 14, 21, and 28 to differentiate recrudescence from reinfection by molecular genotyping.

Confirmation of recurrent cases of *Plasmodium* parasites

The identification of *Plasmodium* species was accomplished using nested PCR with species-specific primers targeting the 18S small subunit rRNA genes of *P. falciparum* and *Plasmodium vivax* [17]. For the primary standard PCR, 5 µl of genomic DNA was used in a 25 µl reaction with genus-specific forward and reverse primers (rPLU5 and rPLU6). For the secondary amplification reaction, 2 µl of primary PCR product was used as template DNA, in which species-specific rFAL1-rFAL2 for *P. falciparum* and rVIV1-rVIV2 for *P. vivax* were used in two separate reactions. The amplified products were electrophoresed on 2% agarose gels. After staining with Gel-red, the gel was visualized under a UV transilluminator, and DNA fragments were estimated using a 100 base pair DNA ladder. All recurrent patients were confirmed to have *P. falciparum* mono-infections.

Genotyping of *msp-1* and *msp-2* genes

Nested PCR genotyping was conducted to distinguish recrudescence from reinfection by pairing dried blood spots (samples collected on day 0 and the day of parasite detection). PCR genotyping of two *P. falciparum* polymorphic genes, merozoite surface protein-1 and 2 (*msp-1* and *msp-2*), was performed according to the WHO protocol [18] for samples from recurrent cases. Two rounds of PCR amplification were carried out. In the primary PCR, primers were designed to amplify the entire genetic locus of the *msp-1* and *msp-2* genes, while the secondary PCR targeted the family-specific alleles of *msp-1* (MAD20, K1 and RO33) and *msp-2* (3d7 and FC27). The amplified products were electrophoresed on 2% agarose gels. After staining with Gel-red, the gel was visualized under a UV transilluminator, and DNA fragments were estimated using a 50 base pair DNA ladder. The results were classified as a new infection if a subsequent occurring parasitaemia in which all the alleles in parasites from the post treatment sample are different from those in the admission sample by greater than 20 bp, and a recrudescence, if at least one allele at each locus is common to both paired samples within a 20 bp, for both *msp-1* and *msp-2* [18].

Treatment and follow-up

Drug treatment was given based on weight according to the revised WHO guidelines [19]. Briefly, participants were treated with the standard six-dose regimen of AL (manufactured by Ipca Laboratories Ltd.) as a tablet of 20/120 mg (Mfd: 12/2021; Exp: 11/2024). The drug was used from the health post which routinely given to *P. falciparum* cases. The drug to the health post was supplied by the government, Ethiopian Pharmaceuticals Supply Agency though proper channel. AL was given twice daily for three consecutive days under direct observation by health extension workers. After receiving an initial dose of AL, the participants were monitored for 30 min to determine retention of the drug in line with the WHO guidelines [10]. Fatty or any foods were not provided to patients. On day 0, participants who successfully received the first dose of AL were given appointment cards containing their name, identification code and the date of the next scheduled visit. They were also given the evening dose of the medication to be taken at home while they were observed by community health workers. Participants were advised to return for treatment on day 1 and day 2. Scheduled follow-up visits were scheduled for day 3, day 7, day 14, day 21, and day 28. On each of the

scheduled days, the participants were screened for parasites using microscopy, and their fever and adverse events were assessed. If participants missed their scheduled visit, they were traced by the assigned home visitor on the same day and brought to the health post.

Study participant withdrawal

Participants who were lost to follow-up, who were infected with *P. vivax*, who were missing doses and who were not willing to continue were excluded from the study.

Classification of treatment outcomes

Treatment outcomes were classified based on WHO guidelines [10] as follows:

Early treatment failure (ETF): the development of danger signs for severe malaria on days 1, 2 or 3 in the presence of parasitaemia; parasitaemia on day 2 higher than day 0 irrespective of axillary temperature; parasitaemia on day 3 with axillary temperature ≥ 37.5 °C; and parasitaemia on day 3 ≥ 25% of the count on day 0.

Late clinical failure (LCF): the development of danger signs for severe malaria in the presence of parasitaemia, the presence of parasitaemia and an axillary temperature ≥ 37.5 °C or a history of fever on any day from day 4 to day 28, without previously meeting any of the criteria of ETF.

Late parasitological failure (LPF) was defined as the presence of parasitaemia on any day from day 7 to day 28 and an axillary temperature < 37.5 °C without previously meeting any of the criteria of ETF or LCF.

Adequate clinical and parasitological response (ACPR): the absence of parasitaemia on day 28, irrespective of axillary temperature without previously meeting any of the criteria of ETF, LCF or LPF.

Assessment of adverse events

Adverse events were assessed through direct questioning and physical examination with a standard list of malaria-associated and AL-related adverse events per WHO guidelines [10].

Data analysis

The data were double entered into the WHO Excel spreadsheet designed for the therapeutic efficacy data. The data were also entered into SPSS version 26 to calculate descriptive statistics (means, standard deviations, percentages and ranges). The cure rate, cumulative success rate and cumulative failure rate were analyzed by using standard WHO per-protocol analysis and Kaplan–Meier survival estimates. The primary efficacy indicator was presence of parasitaemia in the study subject after the start of AL within 28 days. In PCR uncorrected cure rate, it was calculated as all the subjects without parasitaemia divided by total subjects who completed the study per protocol. Whereas, in PCR corrected cure rate, it was calculated as all subjects without parasitaemia divided by total subjects who completed the study per protocol by excluding new infection [10].

Ethics approval and consent to participate

This study was approved by the Institutional Research Ethics Review Board of Arba Minch University (Ref. No: IRB/1293/2022). Permission letters were obtained from the relevant offices. Consent to participate in the study was obtained from each participant.

Results

Baseline characteristics, enrollment and follow-up of study participants

A total of 452 patients suspected of having malaria visited health posts during the study period (Fig. 2), of which 89 *P. falciparum*-monoinfected patients were recruited for the study. Out of the 89 recruited study participants, 72 completed the therapeutic efficacy study (Fig. 2).

A total of 89 patients were recruited for the study; 57.3% (51/89) were males, and 42.7% (38/89) were females. The mean age of the study participants was 25.1 years, and their mean body temperature on day 0 was 37.55 °C. Among the study participants, 65.2% (58/89) had a fever with a body temperature ≥ 37.5 °C, while the remaining 34.8% (31/89) reported having fever within the previous 24 h. Among the study participants, 11.2% (10/89) tested negative for RDT, but *P. falciparum* was detected through microscopic examination and nPCR. At baseline, the gametocyte carriage rate was 13.5% (12/89). The overall baseline geometric mean parasitaemia was 21494, and gametocytaemia was 344 per µl of blood (Table 1).

Adverse events

No adverse events were reported throughout the 28 days of follow-up.

Fever, asexual parasite and gametocyte clearance rate

The study participants were 100% free from fever, asexual parasites and gametocytes on days 2, 3, and 14, respectively (Fig. 3).

Recurrent malaria

During the days of follow-up, five participants experienced a recurrence of parasitaemia (one participant on day 14 and the remaining on day 21). On the day of presentation, all these patients were fever-free.

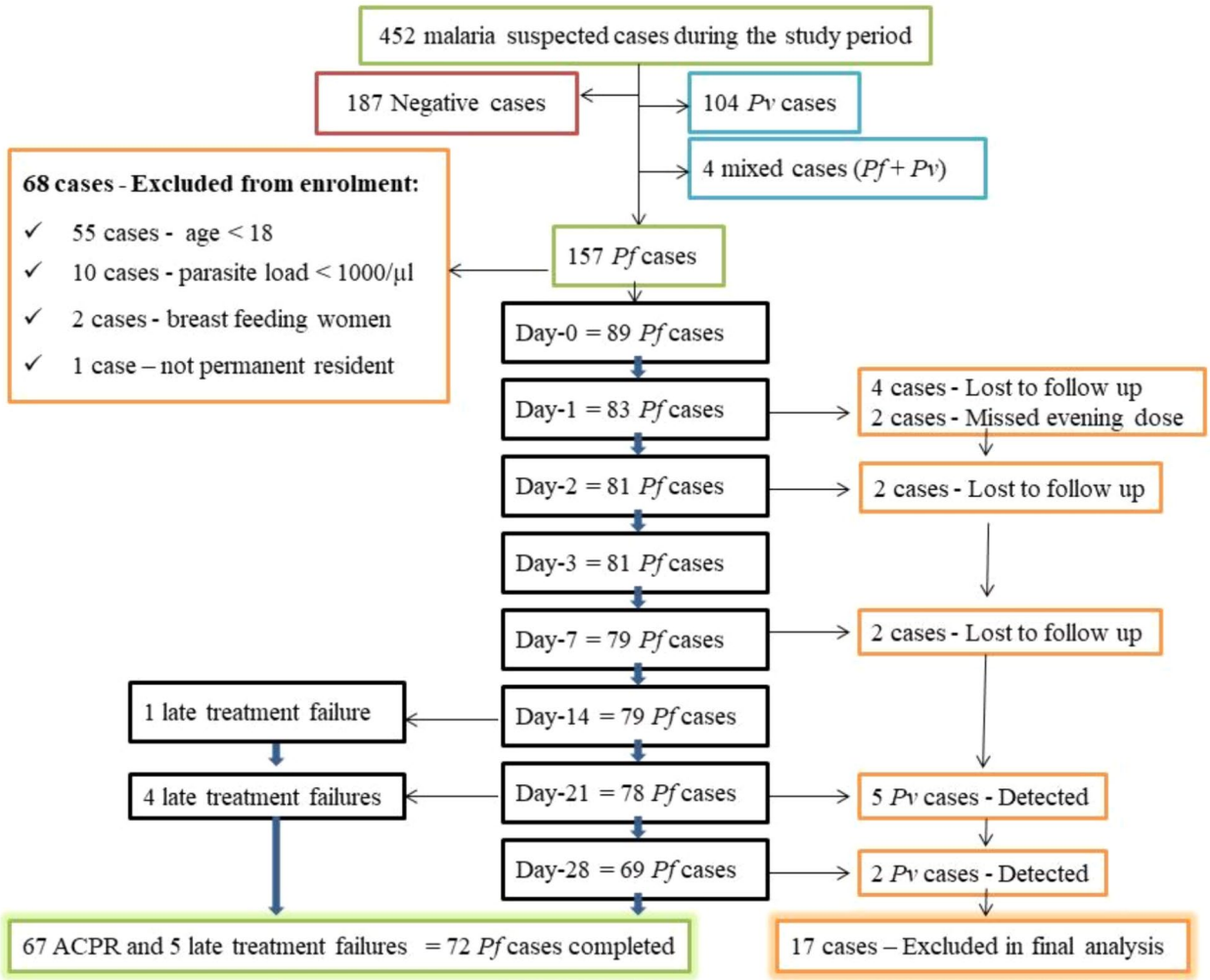

Fig. 2 Flow chart showing the enrollment and follow-up of study participants for the AL efficacy study at Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia, July–October 2022 (*Pf*: *P. falciparum*, *Pv*: *P. vivax,* and ACPR: Adequate clinical and parasitological response)

Table 1 Baseline sociodemographic and clinical characteristics of the study participants at Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia, July–October 2022

Characteristics	Measured values
Sex (Male), n (%)	51 (57.3)
Sex (Female), n (%)	38 (42.7)
Mean age in year (range)	25.1 (18–43)
Mean weight in kg (range)	59.1 (43–72)
Mean axillary temperature in °C (± SD)	37.55 (± 0.74)
Febrile participants (≥ 37.5 °C), n (%)	58 (65.2)
RDT negative n (%)	10 (11.2)
GM parasitaemia/µl (range)	21,494 (1120–196000)
Gametocyte carriage n (%)	12 (13.5)
GM gametocytaemia/µl (range)	344 (80–3200)
Total n (%)	89 (100)

SD: standard deviation; kg: kilogram; GM: geometric mean

However, two of them had experienced fever in the previous 24 h. Three participants were classified as recrudescent failure after molecular analysis. At least one common allele was detected within a 20-base pair interval by *msp-1* and *msp-2* genotyping on day 0 and on the day of recurrence. On the other hand, the other two recurrent participants were classified as having reinfections since they had different parasite strains on day 0 and on the day of recurrence (Table 2).

Cure rate of AL

Based on the per-protocol analysis, the PCR-uncorrected cure rate of AL among the study participants was 93.1% (67/72) (95% CI 84.5–97.7), while the PCR-corrected cure rate was 95.7% (67/70) (95% CI 88.0–99.1). Five treatment failures were observed (2 LCFs and 3 LPFs). In this study, no ETFs were detected (Table 3). Based

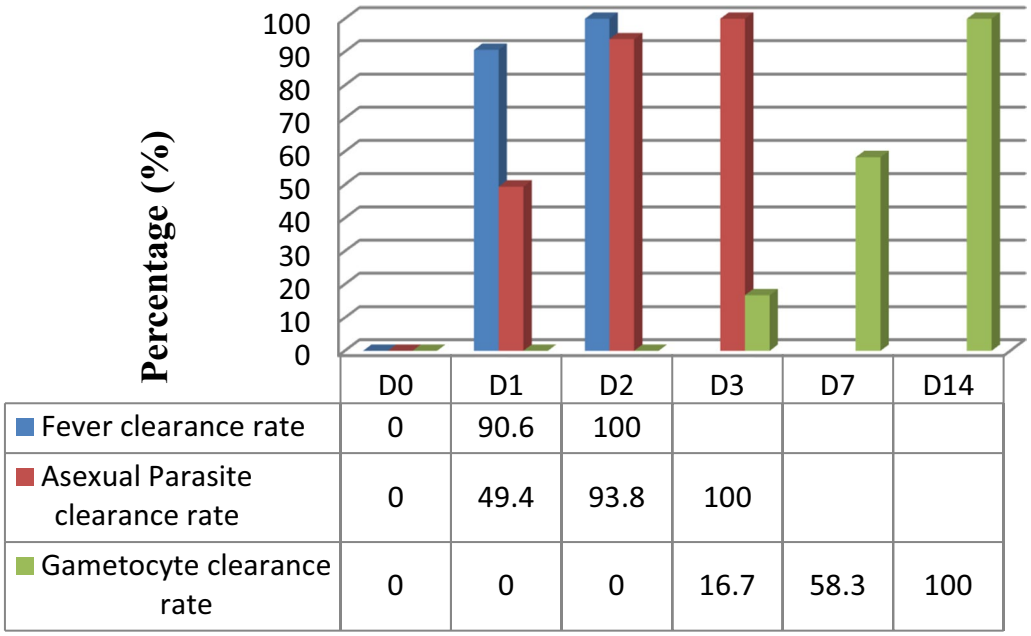

Fig. 3 Fever, asexual parasite and gametocyte clearance rates among the study participants in Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia, July–October 2022

Table 2 Estimated amplicon size (bp) of the *msp1* and *msp2* alleles for participants with recurrent *P. falciparum* infection on day 0 and day of recurrence in Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia, July–October 2022

	Sample ID	*msp1*			*msp2*		Results
		MAD20	K1	R033	FC27	3D7	
Participant A	09d0	–	100, 190	–	400	–	Recrudescence
	09d21	200, 250	130, 200	180	450	–	
Participant B	35d0	200, 250	200	180	300,400	400	Recrudescence
	35d21	250	200	180	290,390	380	
Participant C	43d0	–	200	200	–	–	Reinfection
	43d21	200, 250	130	100	–	–	
Participant D	61d0	250	200	100,180	400	300	Reinfection
	61d14	220	100	–	–	–	
Participant E	77d0	250	200	180	–	480	Recrudescence
	77d21	–	190	100	250,400	–	
Allele size (bp)		160–250	100–200	75–200	250–450	300–500	Correct band size

on Kaplan–Meier survival analysis, the PCR-corrected cumulative incidence of AL success rate was 93.7% (95% CI 85.5–97.3), and the PCR-corrected cumulative incidence of AL success rate was 96.2% (95% CI 85.5–98.7) (Table 3).

Discussion

This study showed that AL has high therapeutic efficacy in treating uncomplicated *P. falciparum* malaria in the study area, with high parasite clearance and cure rates. The study also revealed that AL was able to clear asexual parasites from patients within three days, gametocytes within 14 days and fever within two days. Although the effectiveness of AL was confirmed in the study population, attention is still needed for cases of recrudescence.

Fever is the main clinical manifestation of malaria and can cause severe discomfort. *Plasmodium falciparum* appears to exploit the innate febrile response to mediate resistance to artemisinin [20]. In this study, all patients were fever-free on day 2, which could be an indication of

Table 3 Summary of treatment outcomes with and without PCR correction among study participants who were treated with AL at Sille Village, Arba Minch Zuria District, Gamo Zone, Southwest Ethiopia, July–October 2022

Efficacy endpoints	n (%)	95% CI
ETF	0	0.0–5.0
LCF without PCR-correction	2 (2.8)	0.3–9.7
LCF with PCR-correction	1 (1.4)	0.0–7.7
LPF without PCR-correction	3 (4.1)	0.9–11.7
LPF with PCR-correction	2 (2.9)	0.3–9.9
ACPR without PCR-correction	67 (93.1)	84.5–97.7
ACPR with PCR-correction	67 (95.7)	88.0–99.1
Total Patients at baseline	89	–
Total patients in PP before PCR-correction	72	–
Total patients in PP after PCR-correction	70	–
PP PCR-uncorrected cure rate	67/72 (93.1%)	84.5–97.7
PP PCR-corrected cure rate	67/70 (95.7%)	88.0–99.1
K–M PCR-uncorrected cure rate	93.7%	85.5–97.3
K–M PCR-corrected cure rate	96.2%	85.5–98.7

ACPR: adequate clinical and parasitological response; ETF: early treatment failure; K–M: Kaplan–Meier method; LCF: late clinical failure; LPF: late parasitological failure; PCR: polymerase chain reaction; PP: per-protocol analysis

the effectiveness of AL in the treatment of *P. falciparum* malaria. The absence of asexual parasites was confirmed in all patients on the third day of microscopic examination, as stated in the above paragraph. This finding is comparable with other findings obtained in Ethiopia [12, 13, 21–23].

Asexual parasites were not detected by microscopy on the third day after *P. falciparum* was treated with AL. This finding aligns with previous studies [13, 24–28] and the nature of AL. Artemether is quickly absorbed, and its active ingredient reaches a high concentration within two to three hours of ingestion [25]. This allows the parasites to be cleared quickly. However, lumefantrine is a slow-acting drug that prevents recrudescence by eliminating any remaining parasites [25]. However, other studies reported the presence of parasitaemia on day 3 [12, 26, 27], which might be due to high levels of baseline parasitaemia, host nutrition and immune status [10] or the presence of drug-resistant parasite strains [6–8].

Gametocytes were detected in a few of the study participants. This could be due to the developmental nature of the *P. falciparum* gametocyte, which requires 9–12 days after the development of asexual parasites [28], and it hides in extravascular sites such as the bone marrow until maturation [29]. In addition, this might also be due to light microscopy, as its gametocyte detection ability is minimal [30].

In the presence of AL, none of the gametocyte-positive cases exhibited gametocyte clearance until day three, and complete gametocyte clearance was detected in all the patients on day 14. This indicates that the effect of AL on gametocytes is slower than its effect on asexual parasites. This highlights the possibility of malaria transmission after complete clearance of the asexual stage. This result is comparable with other findings obtained in Ethiopia, Kenya and Zambia [22, 23, 27, 31, 32], which showed complete clearance of gametocytes on day 7 and the presence of gametocytes on day 14 and even until day 42.

Based on the nPCR genotyping, three patients were confirmed to experience recrudescence, and the remaining two were confirmed to experience reinfection. Reinfection is expected in malaria-endemic areas [33]. However, these cases of recrudescence could be explained by factors associated with AL treatment failure, such as the presence of resistant strains [34, 35], poor drug quality [36], poor host nutritional status [37] and incomplete drug metabolism [38].

The PCR-corrected cure rate of AL in this study was 95.7%, which showed the high therapeutic efficacy of AL against uncomplicated *P. falciparum* malaria. Within the range of WHO recommendations, the cure rate of AL for *P. falciparum* malaria should be at least 90% [10]. The observed PCR-corrected cure rate of AL in the current study is comparable to that reported in other studies conducted in different parts of Ethiopia [12, 13, 21–24] and elsewhere in Africa [26, 39–41].

Studies have shown that RDT has better sensitivity than microscopy [42]. However, in the present study, 11.2% of the study participants were RDT negative but microscopically positive for *P. falciparum* malaria. This is in line with studies conducted in Ethiopia [17, 18] and other countries [43]. Such cases might be linked to *pfhrp*2/3 deletion [44]. The reported false negative RDT percentage is high, which demands a change in RDT [24].

In this study, the genotyping of strains did not include glutamine-rich protein (*glurp*), which is commonly used together with merozoite surface proteins (*msp*) 1 and 2. The resistance test was not performed for recrudescent cases, and parasite clearance was not confirmed by PCR, whereas microscopy may miss some cases. In addition, urine test was not performed for pregnancy. The study relied on participants' pregnancy report. Therefore, these factors could be considered as limitations of this study.

Conclusion

The therapeutic efficacy of AL is high in the study area. Therefore, the continuation of AL as a first-line treatment for uncomplicated *P. falciparum* malaria is possible. However, the detection of recurrent patients highlights the necessity of continuing to study the efficacy of AL

treatment. In addition, more than 10% of *P. falciparum* cases were not detected by RDT, which indicates the need to evaluate the performance of RDT in the study area.

Abbreviations

ACPR	Adequate clinical and parasitological response
ACT	Artemisinin-based combination therapy
AL	Artemether–lumefantrine
CI	Confidence interval
ETF	Early treatment failure
K–M	Kaplan–Meier
LCF	Late clinical failure
LPF	Late parasitological failure
PCR	Polymerase chain reaction
PP	Per-protocol
RDT	Rapid diagnostic test
WBCs	White blood cells
WHO	World Health Organization

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12936-024-05087-7.

Supplementary Material 1.

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Author contributions
D.D. involved in study design, data collection, laboratory sample analysis, data analysis, interpretation of the results and drafted the manuscript. B.W., D.W. and F.M. were involved in study design, supervision, data analysis and interpretation of the results. L.G. was involved in data analysis and interpretation of the results. G.S.A., G.T. and Z.Z. were involved in the laboratory sample analysis and results interpretation. Z.Z. and T.M. were involved in data collection and data analysis. B.W., D.W., F.M. and L.G. revised the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials
All data supporting the findings of this study are available within the paper and its Supplementary Information.

Declarations

Ethics approval and consent to participate
This study was carried out following ethical approval obtained from the Research Ethics Review Board of Arba Minch University (Ref. No: IRB/1293/2022). Written informed consent was obtained from each study participant.

Consent for publication
All the authors have read the final manuscript and provided consent for publication.

Competing interests
The authors declare no competing interests.

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