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Description of Serotype 3 Dengue Fever Virus: Clinical, Surveillance and Geographical Expansion in the Northeastern Ethiopia, 2023

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Abstract: Dengue fever (DF) is emerging a mosquito-borne viral infection that commonly found in tropic and subtropic regions. It is caused by dengue virus, which is transmitted to human through the bites of infected with female *Aedes* mosquito during in the day time. Symptoms of DF includes high grade fever, headache, joint and muscle Paine, rash, and milled bleeding. dengue virus contains 4 different serotypes (DENV-1, DENV-2, DENV-3, and DENV-4). On April 4, 2023, a dengue fever (DF) outbreak in the Afar region was detected. From April 4 through June 26, 2023, there were over 6,133 cases reported, along with 4 facility Death were recorded. One hundred twenty tow cases (1.99%) were treated and managed in the inpatient department (IPD) and 64 cases had lab confirmation using RT-PCR from 154 suspected cases from the seven districts with the positivity rate was 41.6%. After 22 days had passed since dengue fever was confirmed in the area, a national response was deployed. Data from a line list compiled by the Afar Public Health and Research Institute from private and public health facilities between April 4 and June 26, 2323, was analyzed. Therefore, the objective of this analysis is to outline and emphasize the effect of DF in Afar, Ethiopia. The total attack rate was 11.91 / 1000 population, while the case fatality rate was 0.6 / 1000 population. By 33.13/1000 population, Semera-logiya town had the most impact compared to the other districts and towns. In this study, men were 54.4% more affected than women. Ages 15 to 24 in both sexes were more affected than older age groups by 33.1%, followed by older age groups 25 to 34 by 29.1%. Fever, which was present in 98.4% of cases, was followed by joint pain (91.5%) while bleeding from orifices was (1.6%).

Keywords: Dengue Fever, Serotype 3, Clinical, Surveillance, Expansion, Northeastern, Ethiopia

1. Introduction

Dengue fever, commonly known as 'breakbone fever' is the most rapidly spreading vector-borne viral infection, transmitted between people by the infected female *Aedes aegypti (Ae)* mosquito [1] while biting during the day time. Globally, the incidence increased by 30-fold throughout tropical and sub-tropical climates worldwide, mostly in urban and semi-urban areas [2].

The infection is caused by one of four dengue virus serotypes (DENV-1, DENV-2, DENV-3, and DENV-4) that belong to the *Flaviviridae* family and belong to the genus *flavivirus*. Pathogens that cause Zika virus also includes pathogens that cause Zika virus disease, yellow fever and West Nile fever [3].

The Symptoms of dengue include fever, muscle and joint pain, severe headache, pain behind the eyes, nausea, rash and vomiting; severe dengue causes severe abdominal pain, persistent vomiting, respiratory distress, organ impairment and death [4, 5]. Other complications of DENV infections such as encephalitis, myelitis and fulminant hepatitis were also reported Recovery from infection is believed to provide lifelong immunity against that serotype [6]. However, cross-immunity to the other serotypes after recovery is only partial and temporary. Subsequent infections (secondary infection) by other serotypes increase the risk of developing severe dengue [7].

The risk factors include rising resistance to insecticides among vectors and also the incidence of the disease may increase with changing climate, continued rapid unplanned urbanization and the abundance of disposable, non-degradable containers that serve as breeding sites in the peri- domestic environment for the principal vector, *Aedes aegypti*, which maintains the urban dengue transmission cycle among humans in most Africa countries [7].

The impact of dengue virus infection in Africa has yet to be determined. Outbreaks of dengue fever and dengue hemorrhagic fever are infrequently recorded; nonetheless, we cannot conclude that moderate and severe dengue infection is uncommon in African countries. The spread of distinct dengue virus serotypes is likewise poorly documented [8]. This is mostly due to a lack of laboratory surveillance systems or their inadequate implementation, which is the source of missing information on dengue virus activity.

The incidence of dengue has increased drastically (by more than 300% since 2009) [9]. According to WHO, 2020 estimated, 3.9 billion people are at risk in 128 countries of which more than 100 are endemic for dengue; the majority of those affected live in Africa, Asia and Latin America [10]. Furthermore, approximately 3 million people with disabilities were diagnosed in the same year [11].

Dengue epidemics have been reported in Africa since the 19th century, in countries including Zanzibar (1823, 1870), Burkina Faso (1925), Egypt (1887, 1927), South Africa (1926–1927), and Senegal (1927–1928) [12]. Between 1960 and 2010, there were 20 laboratory-confirmed outbreaks in 15 African nations, with the majority occurring in Eastern Africa. All four-dengue virus (DENV) serotypes have been identified in Africa, with DENV2 generating the most severe epidemics [12].

DENV-3 isolated during epidemics in Puerto Rico in 1963 [13] and has been frequently associated with severe dengue outbreaks in Asia, Africa and Latin America [8, 14-17]. The first report of DENV-3 activity in Africa was linked to outbreaks in Pemba, Mozambique, in 1984 and 1985, with two deaths from dengue Haemorrhagic fever [18]. After ten year later DENV-3 was then detected in 1993 in Somalia and areas around the Persian Gulf [19]. The after these, DENV-3 circulation in West Africa was first identified in a traveler returning to Spain from Cameroon in 2006 [7].

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In Africa, dengue has been reported in 34 countries, mostly in Eastern Africa. In countries bordering Ethiopia, such as Sudan, Eritrea, Kenya and Djibouti, Dengue has been often reported. In Ethiopia, the first major outbreak occurred in Dire Dawa city (East Ethiopia), with a total of 11,409 cases [20]. The following year, in 2014, another outbreak was reported in Dire Dawa, as well as in Godey Town, Somali Region (South-East Ethiopia) and in Ada'ar Woreda, Afar Region (North Ethiopia) [21, 22]. Subsequently, annual outbreaks have been observed in Godey Town and Dire Dawa [23, 24]. During the response to the covid 19 pandemics, the dengue fever outbreak continued in Dira dawa city and the Ethiopian Somali region warder districts and Gewane District, Afar Region, northeastern Ethiopia [25-27]. Most outbreak investigations thoroughly investigate the risk factors of dengue fever outbreaks. However, with the exception of a study conducted in Dire Dawa in the year 2020, only serotype 1 was found, while the other investigations did not identify the serotype of dengue fever. Even a study at Metema and Humera hospitals in northwest Ethiopia from 2016 to 2017 found dengue fever virus in febrile patients who visited the two facilities [28] but did not clearly explain the serotype.

The Afar regional public health and research institute notified the Ethiopian Public Health Institute (EPHI) / Public Health Emergency Management (PHEM) of the first probable DF outbreak in Mille Town Zone 1 in Northeastern Ethiopia on April 4, 2023, and requested an investigation.

Hence the objective of the study is to provide epidemiologic data on dengue fever in the Afar region that will serve as a baseline for monitoring disease occurrence trends and identifying epidemic impacted areas. Furthermore, trend analysis of incidence could assist public health officials in enhancing vector management to reduce the burden of the disease.

2. Materials and Methods

Study Setting: The Afar Region formerly known as region 2, is a regional state in northeastern Ethiopia. Its capital is the planned city of Semera, which lies on the paved Awash–Assab highway. It's bordered by Eritrea to the north and Djibouti to the northeast, and shares regional borders with the Tigray, Amhara, Oromo and Somali regions.

The administrative system of the region is divided into five administrative zones: Awsi Rasu (Zone 1), Kilbet Rasu (Zone 2), Gabi Rasu (Zone 3), Fanti Rasu (Zone 4), and Hari Rasu (Zone 5) as well as 37 woredas and five city administrations. Semera Logiya is the capital city of the region and is located in Zone 1; additional woredas and towns in Zone 1 include Ada'ar, Afambo, Asayita, Chifra, Dubti, Elidar, Kori, and Mille.

According to the 2012 (EFY) Health and Health Related Indicators published by Federal MoH [29], Afar has 7 Hospitals, 96 Health Centers and 338 Health Posts. According

to the Central Statistical Agency (CSA) of Ethiopia, the Afar Region has a total population of 1,945,801 people, with 832,650 men and 661,549 women. According to the CSA, rural residents make up 86.6% of the population, while urban residents make up 13.4% [30].

This region has an estimated area of 72,052.78 square kilometers and a population density of 20.7 persons per square kilometer. While, recurrent drought, disease outbreaks, locust infestation and conflicts affect the Afar communities and also Chronic water shortage in the region also forces the population and their livestock to move from place-to-place affecting children's education, health, and wellbeing.

Data collection and Analysis: line list data was used following the declaration of the dengue fever outbreak in the Afar's region, Zone one, Mille district. The data was collected in the aftermath of the DENGUE FEVER epidemic report. Health facilities (Hospitals, Health centres and private health clinics and hospitals) were visited to check whether all information required is filled on the line list as of coming patients and Microsoft Excel sheet 2021 was used to clean and check the data for competence before exporting it to SPSS 25 statistical software for analysis. It is further described in terms of person, place, and time. Rate, ratio, and proportion were calculated using descriptive analysis for the parameters of interest. Narrations, frequency distributions, line graphs, pie charts, bar graphs, and histograms were used to show the results.

Case definition of dengue fever:

The suspected case definition of dengue fever typically includes individuals who present with the following criteria:

1. Acute febrile illness: the person has an acute onset of high-grade fever (usually lasting 2-7 days).
2. Two or more the following symptoms: headache, sever joint and muscle pain, pain behind the eye, rash, nausea/vomiting or unusual bleeding or bruising.
3. Exposure history: the person has a recent travel history to residence in an area where dengue fever is known endemic.

The probable case definition of dengue fever typically includes a combination of clinical symptoms and epidemiological factors.

1. Clinical symptoms: Acute onset of high fever (usually lasts 2-7 days), severe headache, often behind the eyes, muscle and joint pain, rash (usually appearing 2-5 days after onset of fever, pain behind the eye, nausea and vomiting, mild bleeding manifestation (e. g., nose or gum bleeding, petechial or easy bruising).
2. Epidemiological factors: Recent travel to or residence in an area where dengue i.e. endemic or circulating or being in an area with ongoing dengue fever outbreak or exposure to confirmed dengue case.
3. The confirmed case definition dengue fever typically involves a combination of clinical symptoms and laboratory conformation. The exact criteria may be varied across the different countries and health organization, but here is a general description:
4. Clinical symptoms: dengue fever is characterized by sudden onset of high fever, accompanied by at least two of the following symptoms: sever headache, pain behind the eye, muscle and joint pain, rash or mild bleeding (such as nose or gum bleeding).
5. Laboratory conformation: to confirm the diagnosis, laboratory test can be conducted and may include: detection of dengue virus through molecular test kits PCR (Polymerase Chain Reaction) OR Serological tests to the detect specific antibodies produced in response to dengue infection. This can be done through tests like ELISA (Enzyme-linked immunosorbent assay) or HI (Hemagglutination inhibition).

2.1. Laboratory Investigation

Five milli liters of whole blood were collected from all suspected patients with active disease (presenting with fever), and after centrifuge of whole blood, then 2 ml of serum was collected and shipped to Ethiopian Public Health Institute (EPHI) Arboviral laboratory for Real time polymerase chain reaction (RT- PCR) to carry out IgM Serology test. Triplex tests for Dengue, Chikungunya, and Zika virus were performed at the national arboviral lab. However, eight of the ten blood samples collected on April 2, 2023 were positive for the Dengue fever virus but negative for the remaining arboviral diseases (Chikungunya and Zika).

2.2. Entomological Investigation

The entomological surveys were prompted by the need to identify the vector(s) responsible for the individual DF outbreaks in the area as soon as possible. We identified and selected clinical cases of DF based on epidemiological and virological documentation for this aim. Estimating the house index (HI) and the Breteau index (BI) yielded entomological risk indicators based on bio-ecological data.

2.3. Ethical Considerations

Permission to conduct study secured from Afar public health and research institute and Ethiopian public health institute, public health emergency management directorate. While the Retrospective nature of study makes no consent was sought from the patients.

3. Result

Clinical and Epidemiological description of dengue fever outbreak: The Ethiopian Public Health Institute (EPHI) received reports of suspected Dengue fever virus cases in the Afar's region, Zone one, Mille districts on April 4, 2023, and 10 blood samples were taken from these suspected patients. The National Arboviral laboratory performed an RT-PCT triplex test for Dengue, Chikungunya, and Zika, and the findings revealed that 80% (8/10) of the cases tested positive for dengue fever with serotype 3 but negative to other Arboviral disease. The test results were forwarded to the Afar Public Health and Research Institute (APHRI) in order to enhance local surveillance and implementing standard case

management for dengue fever cases. On top of these activities various disciplines of outbreak investigation teams, including medical practitioners, Epidemiologists, Laboratory personnel, and Entomologists, were deployed to the afflicted district to investigate and providing prompt outbreak response. This was after 22 days of the dengue fever outbreak being confirmed. The team developed a line list form and carried out case registration for suspicious patients, as well as community sensitization and capacity building for health care practitioners, and an alert letter was sent to Mille town's neighboring districts and towns. After a week of sending letters to districts and towns, identical sign and symptoms were reported from other districts and town called Semera-logiya, and the national arboviral laboratory confirmed that it was dengue fever. From April 4th, 2023 to June 22nd, 2023, the region registered and reported 6,133 suspected and confirmed cases in seven towns including Aminbara, Awash, chifra, Gelealo, Gewane, Mille, and Semera-Logiya in the line list (Figure 3). On April 22/2023, the highest number of cases reported per day was 185, and the most afflicted area was Semera Logiya City, with 2,961 cases, followed by Mille Town 1933 cases.

From April 2, 2023, to May 26, 2023, 154 laboratory cases were collected in seven districts, and 64 cases were confirmed by Reverse Transcription Polymerase Chain Reaction (RT-PCR) at the national rabies laboratory, resulting in a positivity rate of 41.6%. From the line list analysis 5,832 (95.1%) were probable and the remaining 237 (3.8%) were suspected. The overall attack rate in the two zones of the Afar region was 11.91/1000 population, with Semera-Logiya town being the most impacted among Zone one districts and towns, followed by Mille town, with attack rates of 33.31 and 15.90/1000 population, respectively (Figure 6). Most the cases were reported from Zone three districts and towns (Figure 7).

Prior to the arrival of the National and Regional of rapid response team (RRT), four facility deaths were reported in the districts (Mille town); in addition, they were discussed with kebele leaders (the smallest administrative unit in Ethiopia) and told us that there were more than five unknown deaths every day, while the number of deaths has now decreased; This is when the team began intervention activities such as identifying potential mosquito breeding sites (figure 1), community sensitization, capacity building for health care professionals and health extension workers, including surveillance of Arboviral disease, case management at hospital and private clinics, public health emergency management, enhanced social mobilization, on-the-job training on Rapid Diagnostic Test (RDT) to laboratory staff, and entomological indices.

Figure 1. *Used tyers repair at Ethiopian Shipping and Logistics service garage, Mille town kebele 4, on April 27, 2023.*

Figure 2. *Temporary mosquito breeding site in Semera-Logiya town, on April 27, 2023.*

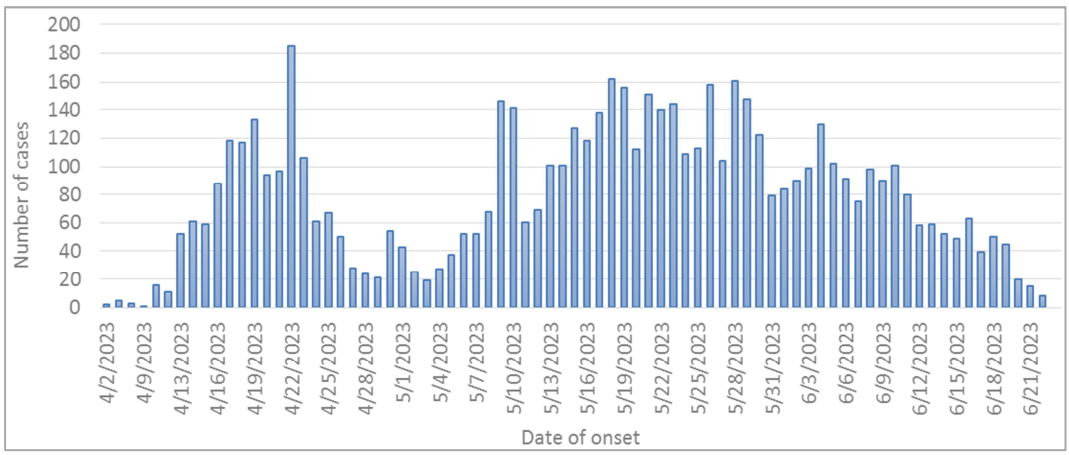

Figure 3. *Distribution of suspected and confirmed dengue fever cases in Afar region during the outbreak course.*

Almost all of the patients Fever (98.6%), joint pain (91%), Headache (89.1%), Chills (84%), Retro orbital pain (74.9%), and Vomiting (44%) were presented in the health facility, while additional significant clinical characteristics such as Bleeding from gum and nose (1.6%) were demonstrated by the patients (Table 1).

Table 1. *Clinical feathers were seen on confirmed and suspected 6,133 DF cases occurrences in Afar from April 4 to June 26, 2023.*

Clinical features	Response	Frequency (n=6133)	Percentage (%)
Fever	Yes	6034	98.4
	No	99	1.6
Chills	Yes	5150	84.0
	No	983	16.0
Joint Pain	Yes	5610	91.5
	No	523	8.5
Vomiting/Nausia	Yes	2749	44.8
	No	3384	55.2
Retro orbital pain	Yes	4596	74.9
	No	1436	23.4
Headache	Yes	5466	89.1
	No	667	10.9
Bleeding	Yes	101	1.6
	No	6032	98.4

3.1. Description by Place

Awsi Rasu (Zone 1) and Gabi Rasu (Zone 3), two zones, recorded occurrences between April 4 and June 26, 2023. Zone three reported 1084 (18%) of the recorded cases, while Zone One reported 5046 (82%) of the cases (figure 4). Gewane districts reported 710 (11.6%) and Gelealo districts 166 (2.7%) cases from Zone Three districts, whereas Semera-logiya districts reported 2962 (48.3%) cases and Mille 1933 (31.5%) cases from Zone One (Figure 4).

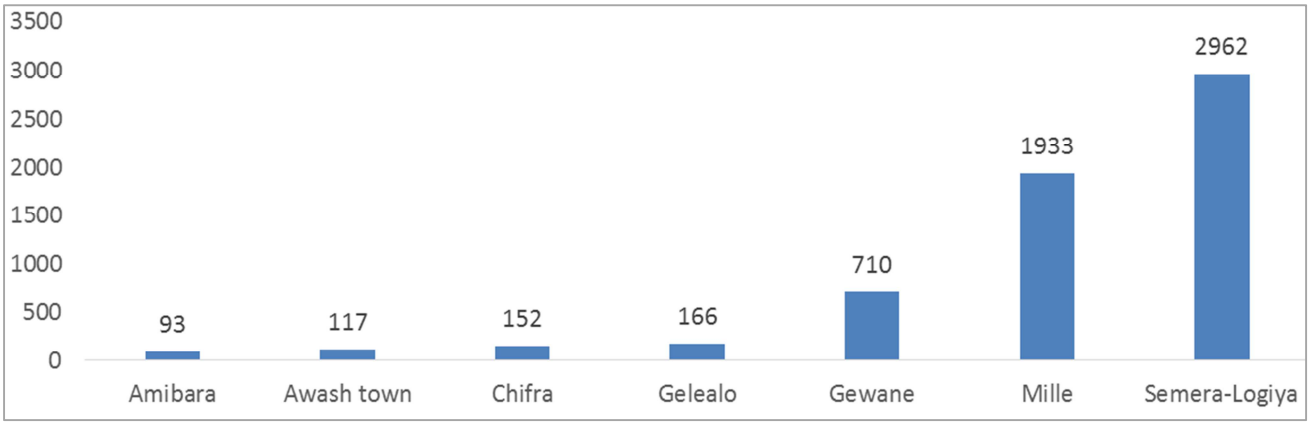

Figure 4. *Dengue fever distribution by districts in Afar region of Ethiopia, August 4 to June 26, 2023.*

Description by Sex and Age: The number of inpatient cases in the seven districts was 122 (1.99%), while the remaining cases, 6,011 (98.1%), were treated as outpatients. There were 3318 (54.4%) men and 2815 (45.9%) females among the 6133 patients shown in table 2. In the present outbreak, Age of patients range from 2 years to 85 years have been documented. However, the majority of cases were between the ages of fifteen and twenty-four (33.1% (2030/6133)), followed by the ages of twenty-five and thirty-four (29.6% (1817/6188)) and age beyond 35 years (18.7% (1145/6133)) in both sexes. The Median age of the current outbreak cases is 12 years and (IQR: 30– 18), as depicted in table 2.

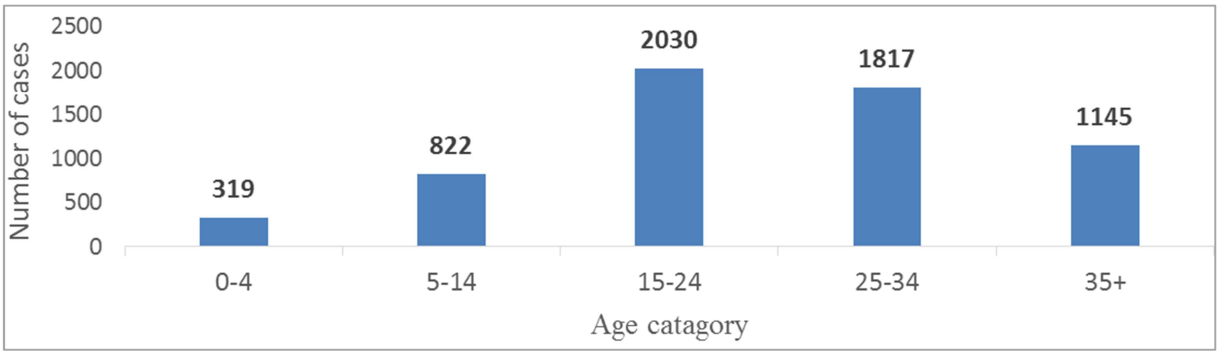

Figure 5. *Dengue fever case distribution by Age group in Afar region of Ethiopia, August 4 to June 26, 2023.*

Table 2. *Cases distribution by Age group and Sex, Afar region of Ethiopia, August 4 to June 26, 2023.*

Age category	Number of cases	percent (%)	Female	Male
0-4	319	5.2	155 (2.5%)	164 (2.7%)
5-14	822	13.4	365 (6.0%)	457 (7.5%)
15-24	2030	33.1	982 (16.0%)	1048 (17.1%)
25-34	1817	29.6	831 (13.5%)	986 (16.1) %)
35+	1145	18.7	482 (7.8%)	663 (10.8%)
Total	6133	100	2815 (45.9)	3318 (54.1)

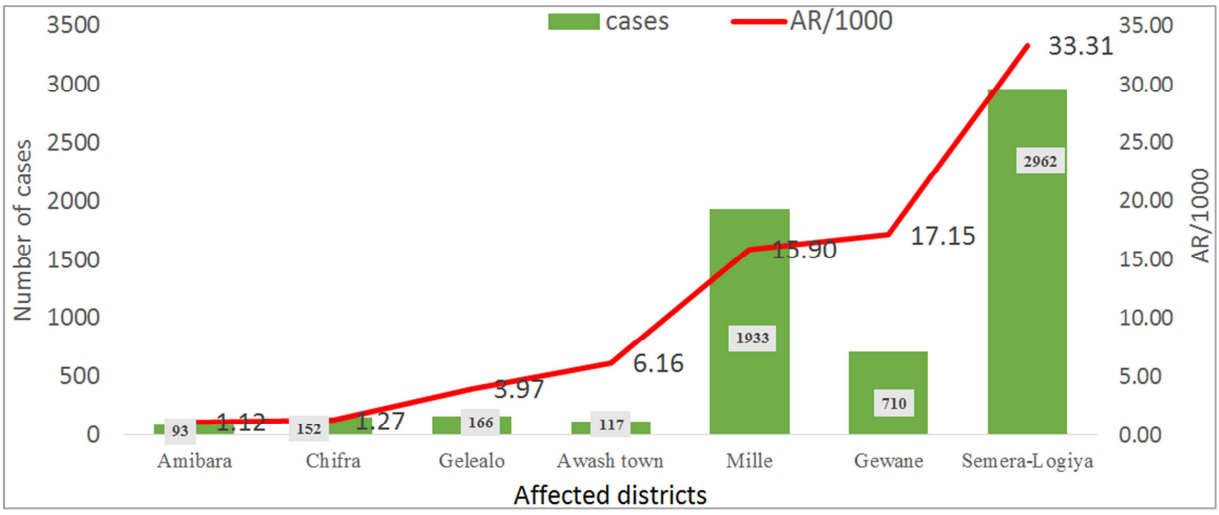

Figure 6. *The Attack Rate of Dengue fever in different districts of Afar region, August 4 to June 26, 2023.*

3.2. Entomological Findings

Aedes aegypti was identified as the vector of the current dengue fever outbreak, and potential breeding sites included old discarded tyers, old rotto, small plastic water holding containers (old Jerican, pot, old cemented jar, and others), and a cemented water holding tank used for hand washing in hotels and cafeterias. According to the Entomological indices, the Breteau Index was 68%, the House Index was 52.1%, and the Container Index was similarly 35.4%.

4. Discussion

The burden of dengue fever virus is not properly recorded and addressed in most African nations due to inadequate surveillance and lab testing capabilities. Dengue fever virus is one of the immediately reportable diseases under the national surveillance system, although the prevalence of dengue fever has been aggressively risen in northeastern and eastern part of Ethiopia. Policy and strategies for preventing and controlling

arboviral diseases are not properly designed or executed at the national level.

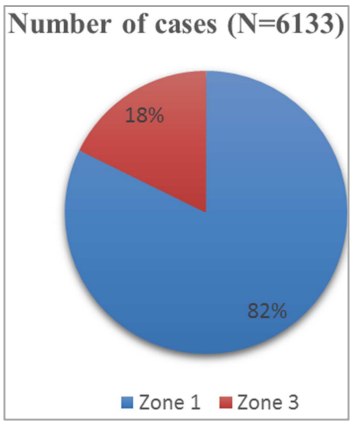

Figure 7. *Case distribution of dengue fever by Zone in Afar region, August 4 to June 26, 2023.*

Ethiopia-Country Commercial Guide report (2019), shown that Ethiopia is one of the landlocked countries and most of the goods essentially transported to and from the port by track using Djibouti ports as main gateway for the international trade. The trade in second-hand tires has been implicated as a factor in this process because they act as mobile reservoirs of mosquito eggs and larvae [31]. Figure 1 depicts the possibility for used tier repair in Mile town and the route from Djibouti, as well as the risk of a massive dengue disease outbreak in the area.

Except for serotypes DENV-4, all other dengue fever serotypes (DENV -1 and DENV-2) have been detected in Dire Dawa city and Ethiopian Somali region in the last ten years [22, 25, 27]. Meanwhile, Dengue fever virus serotype 3 (DENV-3) has been identified and reported in all seven districts and towns of Afar region dengue fever patients since April 4, 2023. The geographical expansion of Dengue fever virus serotype 3 was rapid. Similar to a study conducted in Pakistan, more patients with the serotype (DENV-3) have been identified compared with other serotypes [32].

During this outbreak, the total population affected was 6,133, with an overall attack rate of 11.19/1000 population. Mille town was the most impacted, with an attack rate of 33.13/1000 population with the cases fatality rate 0.7/1000 population. The epidemiological parameters of this outbreak were similar with previous research, which also found a higher proportion of DF infections (54.4%) among men due to gender, cultural, and behavioral disparities [25, 26, 33]. The *Aedes* mosquito bites during the day, and in the study area, most females were dressed in long slaves due to Islamic dressing cultures, which may be one of the barriers to mosquito biting, whereas most male individuals were dressed in short slaved t-shirts as results it confirmed that daily wearing of short sleeve T-shirts was strongly associated with a higher likelihood of DF infection compared to those in full dress. These finding similar to other studies conducted in different area [22, 23, 25, 26, 34-38] in other round the male individuals more affected may be gathered under the tree for various purposes such as group discussion and also due to local hot environmental conditions.

According to the findings of this study, the median age of the patients was 12 years old (IQR:18-30), with the age group 15-24 years old accounting for the highest proportion of cases (33.1%). This is similar to previous studies conducted in Dire Dawa, Ethiopian Somali area, where the median age of the cases was 25 years old (IQR: 17-38) and 20 - 24 years old (23.8%) with a median age of 27 years old (IQR: 22-38) [25, 33]. Another study on the epidemic investigation in Mozambique found a median age of 28 years (IQR: 18-35) (IQR: 18–35) [39]. This could indicate that they are active age groups who spent the most of their time outside, exposing themselves to day bite mosquitos. This adult predominance may be attributed to the involvement of adult groups in outside activities or economic activities.

In the descriptive analysis, the admission rate of the patient was 1.99% and the positivity rate also 41.6%. The results similar to the study conducted in Dire Dawa city on Detection of serotype 1-Dengue fever outbreak in Dire Dawa city [25].

Aedes aegypti served as the primary vector in the majority of dengue fever outbreaks, and the most frequent breeding site was old discarded tyers, old Rotto, small plastic water holding containers. Study conducted in Dire Dawa, containers such as tires, flowerpots, Jerry cans (Rotto), were infested by the *Aedes aegypti* mosquito [23, 25, 26, 40]. *Aedes aegypti* breeds in tyers, ground pools, abandoned pots, plastic rubber, buckets, clean water nylons, and aluminum containers, according to studies carried out in Tanzania and Kabridahar Town, Ethiopian Somali region. [22, 41].

5. Conclusion

Dengue fever incidence has increased in the country over the past 10 years, and the fatality rate from dengue fever has also increased with each outbreak episode. This could be due to cocirculations of distinct serotypes of dengue fever virus or to the circulation of different serotypes of dengue fever virus in a similar geographic area. Even if dengue fever virus illness is one of the most immediately reportable arboviral diseases in the country, if facility surveillance and localized response actions are not adequately addressed, the incidence of dengue fever will spread to other areas, claiming the lives of millions. Additionally, in locations where an outbreak has occurred, an investigation should be done. As results strengthen community and facility-based surveillance and lab identify serotype and prompt public response, including active case search, community sensitization, capacity building, standard case management, and the use of integrated vector management critically important.

6. Limitation

Several constraints were recognized and highlighted in these circumstances while analyzing the outcome retrospective data. The analysis may underestimate the true burden because it is based primarily on patients sought at a health facility, leaving out asymptomatic instances in the

community. Recurrent Dengue fever virus outbreaks have been observed in the northern and eastern parts of the country, and while clinical case detection and notification remain a challenge for the country, this is being hidden in some areas by malaria and other acute febrile illnesses. The recurring viral circulation in the area may aggravate mortality and morbidity. The other challenge is providing a prompt and adequate response in affected communities, which is usually too late in most outbreak responses. The other important point in the limitation of dengue fever clinical case management and detection in the country level. The dengue fever virus is still circulating in the community throughout the country, but the economic and social consequences of dengue fever are not fully recognized. As a result, a nationwide serological survey is crucial and required to understand and scrutinize the burden and implications of dengue fever at the national level.

Conflict of Interest

Authors do not have any conflicts of interest in this work.

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