Addis Ababa University, Collage of Health Science, School of Public Health

Ethiopia Field Epidemiology Training Program (EFETP)

Compiled Body of Works in Field Epidemiology

BY

Getachew Abichu (DVM)

Submitted to the School of Graduate Studies of Addis Ababa University in Partial Fulfilment for the degree of Masters of Public Health in Field Epidemiology

June, 2019

Addis Ababa, Ethiopia

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Fufa Abuna (DVM, MVSC, PhD)

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Approval by Examining Board

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Acknowledgements

I would like to thank my mentors Pro. Fikre Enkusilasie and Dr. Fufa Abuna for their continuous support and guidance for all my outputs. I also want to sincerely appreciate the contributions made during outbreak investigation, health profile, and surveillance system evaluation by FMOH, Ethiopian Public Health Institute, Oromia Regional Health Bureaus and District Health Offices and the community members, I would like to give my thanks to academic program (EFETP).

Finally I would Appreciate National Animal Health Diagnostic and Investigation Center (NAHDIC) for continuous logistic and financial support from sample collection to reagents and consumable supply for the end result of animal Brucellosis prevalence study and Anthrax detection and confirmation in Molecular diagnostic laboratory.

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LIST OF ABBREVIATIONS AND ACRONYMS

AR	Attack rate
ARI	Attack rate Index
BSL-3	Biosafety Level Three
CAP	Capsular Antigen
CDC	Center for Disease Control
CSA	Central statistical Agency
DERF-W	Daily Epidemic Reporting Format-Woreda
DNA	Deoxyribose Nucleic Acid
ELISA	Enzyme linked Immunosorbent Assay
EPHI:	Ethiopian public health institute
EPI	The expanded program on Immunization
FMOH	Federal Ministry of health
HEW	Health Extension workers
IPC	Internal Positive Control
LLIN	Long Lasting Insecticidal Net
MCV1	Measles containing vaccines
MIC	Microscopic
NGO	Non-Government Organization
NSP	National strategic plan
P. F.	Plasmodium falciparum
P. M	plasmodium malaria
P. V.	Plasmodium vivax
P.O.	Plasmodium ovale
PA	Protective antigen
PAS	peasant associations
PBS	Phosphate buffer solution
PCR	Polymerase chain reaction

PHEM	Public Health Emergence Management
PVP	Predictive value positive
RDT	Rapid Diagnostic Test
SIAs	Supplementary Immunization activities
SPR	Slide positive Rate
WHO	world health Organization

OUTLINE OF COMPLIED BODY OF WORK

Ethiopian Field Epidemiology Training Program (EFETP) is a two years in-service training program adapted from the United States Centers for Disease Control and Prevention (CDC). The program is designed to assist the Federal Ministry of Health (FMOH) and Federal ministry Livestock and Fisheries (MOLF) as one health approach to track epidemic and zoonotic diseases which confront the public health. This program strengthen early detection of disease outbreaks forming a bridge among human and animal health profession to control public health importance diseases.

This compiled body of work composed of mainly seven main section which performed during residence I and II. Main section includes: outbreak investigations, surveillance data analysis, surveillance system evaluation, health profile assessment, scientific manuscript, project proposal and one research conducted on zoonotic disease.

This document is organized in eight chapters:

Chapter One: Disease outbreak investigation which contain two outbreaks: one with outbreak investigation of Measles in Mieso district, West Hararge zone, Oromia region and the second one was Anthrax outbreak investigation and confirmation in Farta and Smada districts, South Gondor, Amhara regional state.

Chapter two: Surveillance data analysis on Malaria in Sebeta district of six year data to capture the outbreak trend from 2012-2017

Chapter three: Public Health profile assessment in one of the special zone of Oromia region which showed tremendous demographic dynamicity in related to different health and health related facilities in 2018.

Chapter four: public disease surveillance system evaluation in Malaria an existing surveillance which is one of the weekly reportable prioritize disease.

Chapter Five: epi project proposal on Brucellosis one of zoonotic disease having economically and public importance disease.

Chapter six: Written Manuscript on the detection and confirmation of Anthrax outbreak investigation

Chapter seven: Serological and molecular detection of brucellosis from camel, cattle goat and sheep from Eastern part of Ethiopia, which explain current prevalence of the disease from livestock mainly in pastoral communities.

Chapter Eight: Abstract for scientific presentation, contains two abstracts of measles outbreak in Mieso district West Hararge zone, Oromia regional state, and Animal and Human Anthrax in Farta and Smada districts of south Gondor, Amhara regional state.

Chapter 1. Outbreak Investigations

1.1: Investigation of Measles Outbreak in Mieso District, Oromia Regional State, Eastern Ethiopia, December, 2018

Abstracts

Introduction: Measles is one of the commonly occurred disease in different part of Ethiopia with various morbidity and mortality rate, as Oromia health bureau received report from West Hararge zone of Mieso district we conduct outbreak investigation confirmation from December 20-25/2018, potential risk factor was identified and control measure was implemented. The main Objectives of the study was to describe the outbreak in place, person and time and to identify the potential risk factor of an outbreak.

Methods: A 1:3 un-matched case to control study were conducted. With 16 case and 48 control, a suspected case of any person with fever and maculopapular from Mieso district from October 10 to December 15, 2018. A line listed record was revised and a structured questioner was made from both case and control, all the record entered and analyzed with excel and epi.info 7.1.0.6and SPSS version 23 software. Bivariate and multivariate logistic regression was used for statistical analysis

Results: A total of 12 suspected and 4 conformed case were reported from October 10 to December 15, 2018, out of which 9 (56.25%) were female, with the mean age of 2.6 year (range from 6 month to 8 year), out of 48 control 34(70.8%) were female the mean age 40.8 years (range from 6 month to 48 years). Out of reported 4 kebeles Deneba hunde Misoma is the highest case reported 9(56.25%). The overall attach rate (AR) is 1.21% per 1000 population, zero case fatality rate. We Identify lower vaccination status **(AOR): 3.297 95% CI (0.97-0.989)**, and mal-nutrition (**AOR):3.854, 95% CI (2.115-63.84)** were significant independent risk factors.

Conclusion: Measles outbreak was identified, low immunization coverage and malnutrition were identified as a potential risk factor. Mop up mass vaccination campaign and the patient of under five years of age were admitted to Asebot hospital for supplementary diet and intensive care.

Key word: case control study, Mieso district, measles

1.1.1. Introduction

Measles is an acute viral illness caused by a virus in the family paramyxovirus, genus *Morbillivirus*. Measles is characterized by a prodromal of fever (as high as 105°F) and malaise, cough, coryza, and conjunctivitis, followed by a maculopapular rash. The rash spreads from head to trunk to lower extremities. Measles is usually a mild or moderately severe illness. However, measles can result in complications such as pneumonia, encephalitis, and death. Approximately one case of encephalitis2 and two to three deaths may occur for every 1,000 reported measles cases.(1)

Measles is a highly infectious, acute airborne viral disease with an infectious period of four days before to four days after rash onset. It has an incubation period of 10–14 days. Measles can be a serious illness with complications including otitis media, pneumonia and encephalitis (2)

The measles mortality reduction strategy adopted by the African Region includes improving routine measles vaccination coverage, providing a second opportunity for measles vaccination through supplementary immunization activities (SIAs), monitoring the impact of vaccination activities through case-based measles surveillance and improving measles case management(3)

Measles is one of the vaccine preventable diseases among the leading causes of under-five child mortality in Ethiopia. The Expanded Program on Immunization (EPI) was started in Ethiopia in 1980 with six antigens including measles vaccine. The measles vaccination coverage has been increasing over the past years in Ethiopia. The national immunization coverage survey in 2012 revealed routine measles immunization coverage of 68.2% in children 12-23 months of age.

In Ethiopia, the implementation of the regional measles mortality reduction strategy started in 2002 The national Expanded Program on Immunization was established in 1980 in Ethiopia and includes the first dose of measles-containing vaccine (MCV1) to be given at or shortly after the ninth month of age. Routine vaccination services are delivered through fixed health facilities, outreach sites and since 2004, in some regions, during ‘‘Child Health Days, ’’ also referred to as Enhanced Outreach Services. Enhanced Outreach Services are conducted every 6 months to provide routine vaccination services, including measles vaccination, supplemental Vitamin A, deworming tablets and nutritional screening for children(5)

1.1.2. Objective:

General Objective: To describe and identify risk factor associated with the Measles Outbreak in Mieso district of West Hararge zone, recommend corrective measures.

Specific Objective:

- To verify existence of outbreak
- To describe distribution of the outbreak by person, place and time.
- To identify the potential risk factors and propose control measure of measles infection in Mieso districts

1.1.3. Material and Methods

1.1.3.1. Study Area and Period: The study was conducted December 20-25/2018 in West Hararge zone, Mieso district which is one of the seventeen districts in West Hararge zone, the district bordered Somali region from North, Bordode from West, Chiro from East of Oromia regional districts. Based on figures from the Central Statistical Agency in 2005, this district has an estimated total population of 10,328 of whom 5,342 were males and 4,986 were females.(6)

1.1.3.2. Study Design: We described the outbreak using descriptive epidemiology and then conducted unmatched case-control study design from December 20-25, 2018.

1.1.3.3. Data Collection methods

Structured questionnaire were used in order to collect basic epidemiological information: symptoms, date of onset of disease, age group affected, Vaccination history, Nutritional status, travel history, contact history with measles case before the onset of an outbreak.

1.1.3.4. Case definition:

PHEM Standard case definition: A suspected case with laboratory confirmation (positive IgM antibody) or epidemiological link to confirmed cases in an epidemic.

Community based case definition: Any person with fever and maculopapular (non-vesicular) generalized rash and cough, coryza or conjunctivitis (red eyes) OR any person in whom a clinician suspects measles.

We conduct an outbreak investigation to describe an outbreak by place person and time, identifying risk factors by observation of the medical cards from line lists from Asebot Hospital, and review of the immunization coverage from the districts after each case of the sample were taken and confirmed. We observed an Individual person of the case and made an interview of each parents and immunization status, individual house, Nutritional status, sanitation of the house including the environmental sanitation, latrine coverage and waste disposal management, vaccine storage area according to the requirement of the proper storage and optimal temperature was check in the health post where the immunization carried out.

1.1.3.5. Dissemination of the results

Inclusion and Exclusion criteria

Inclusion Criteria

Cases

Any Person who were living in the Mieso district as a family with those patient and near to the cases who had symptoms of measles (generalized Maculopapular rash and fever plus one of the following: cough or coryza (runny nose) or conjunctivitis (red eyes) from October 10 – December 15, 2018 who/ the parents willing to participate in the study was included.

Controls

Any person who were living in Mieso district who/parent was a neighbor to a case and who did not developed signs and symptoms of measles and agreed to participate was included.

Exclusion criteria

Cases

Those who refused to participate or were unconscious were excluded.

Those who were not living in the area/temporary stay were excluded.

Controls

Those who refused to participate were excluded as well as family members from the same household.

1.1.3.6. Data Processing and Analysis: The data were entered and analyzed using Epi-Info7 ™ version 7.1.0.6. Results were presented using graph and table. Attack rate and case fatality rate were also calculated.

1.1.4. Results:

1.1.4.1. Description Epidemiology

Totally we identified 12 suspected and 4 confirmed measles case in four kebeles. According to the district administration report, Measles vaccination status from cases was 3.19 % unknown, 5.31% was vaccinated only one dose and 8.50% was not vaccinated at all which is much lower than standard. A measles case in the district is high in people who are not vaccinated and complete the whole vaccine, there is repeatedly outbreak with in the district from the previous consecutive years.

The case was happened in four Kebeles who were very close to Asebot Hospital and all those Kebeles were adjacent to each other, out of those kebeles Deneba hunde Misoma is the highest case occurs (75%) of the total case.

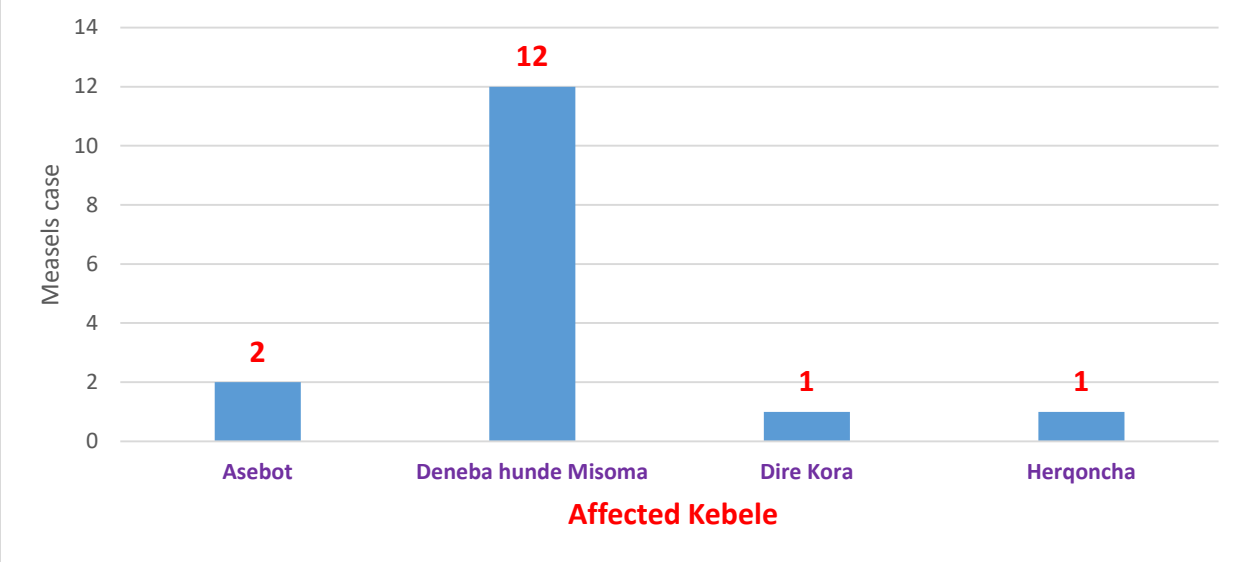

Figure 1: Distribution of Measles case by place in Mieso district, West Harage zone Oromia

Region, October-December 2018.

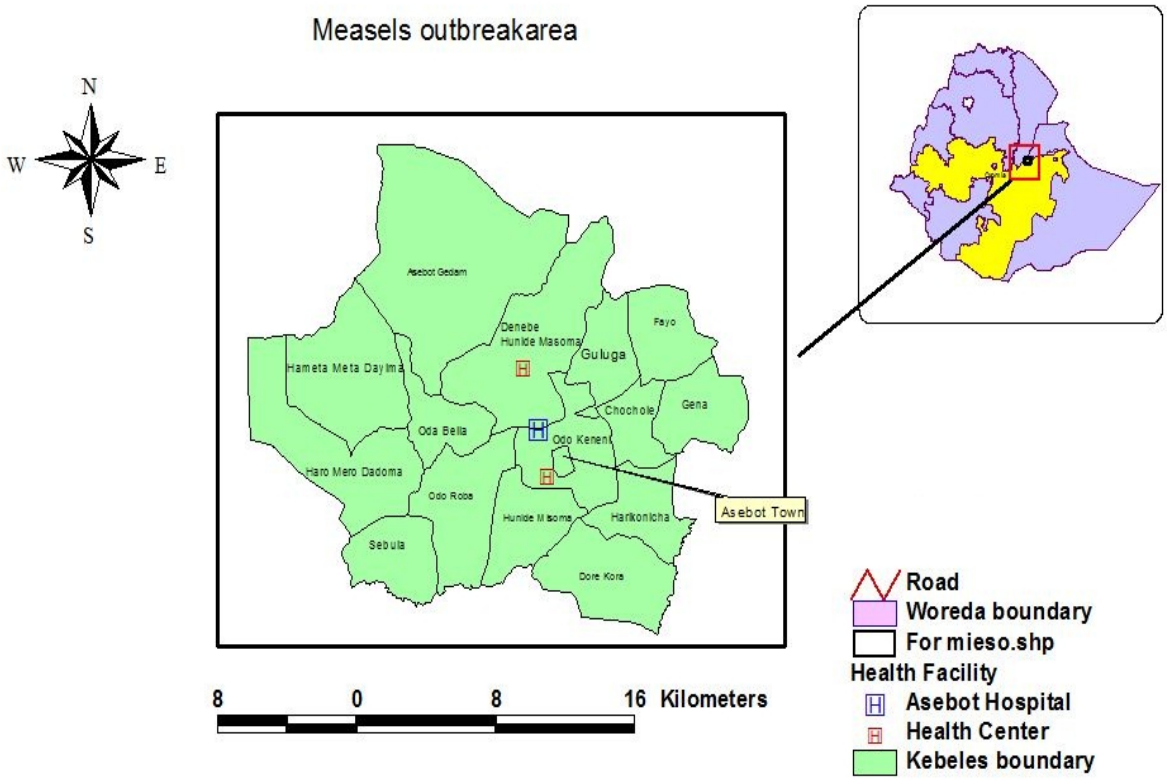

Figure 2: Measles outbreak area Mieso district, West Hararge zone, Oromia regional state, 2019

A total of 12 suspected and 4 confirmed measles cases were identified from October 2 to December 8, 2018. Out of total cases 9(56%) were in Female with the mean age of 2.6 year (range from 6 month to 8 year), out of 48 control 34(70.8%) were female the mean age 40.8 years (range from Six month to 48 years).

Table 1: Attack Rate in Percentage of Measles Outbreak West Hararge Zone Mieso District, Oromia Region 10, October to 15, December, 2018.

Variable	No of cases	Population	AR/1000 population
Age group			
<1yr	2	2345	0.85
1-5yr	12	3589	3.34
6-10 yr.	2	5678	0.35
Sex			
Male	7	6,693	1.04
Female	9	6,444	1.4
Total	16	13,137	1.21
Kebeles			
Deneba Hunde misoma	12	6,032	2
Harqoncha	1	5,044	0.2
Asebot	2	13,137	0.2
Dire kora	1	6383	0.2

The onset of an outbreak was from early of October (10/10/18) to mid of December (12/15/2018)which reported for about 2 months, the last report was which is zero report for 30 days.

Characteristics of case and Control.

West Hararge zonal PHEM officer and Mieso district identified the index case of the measles outbreak. An index case was Female of 8 years old who started to attend her primary school after

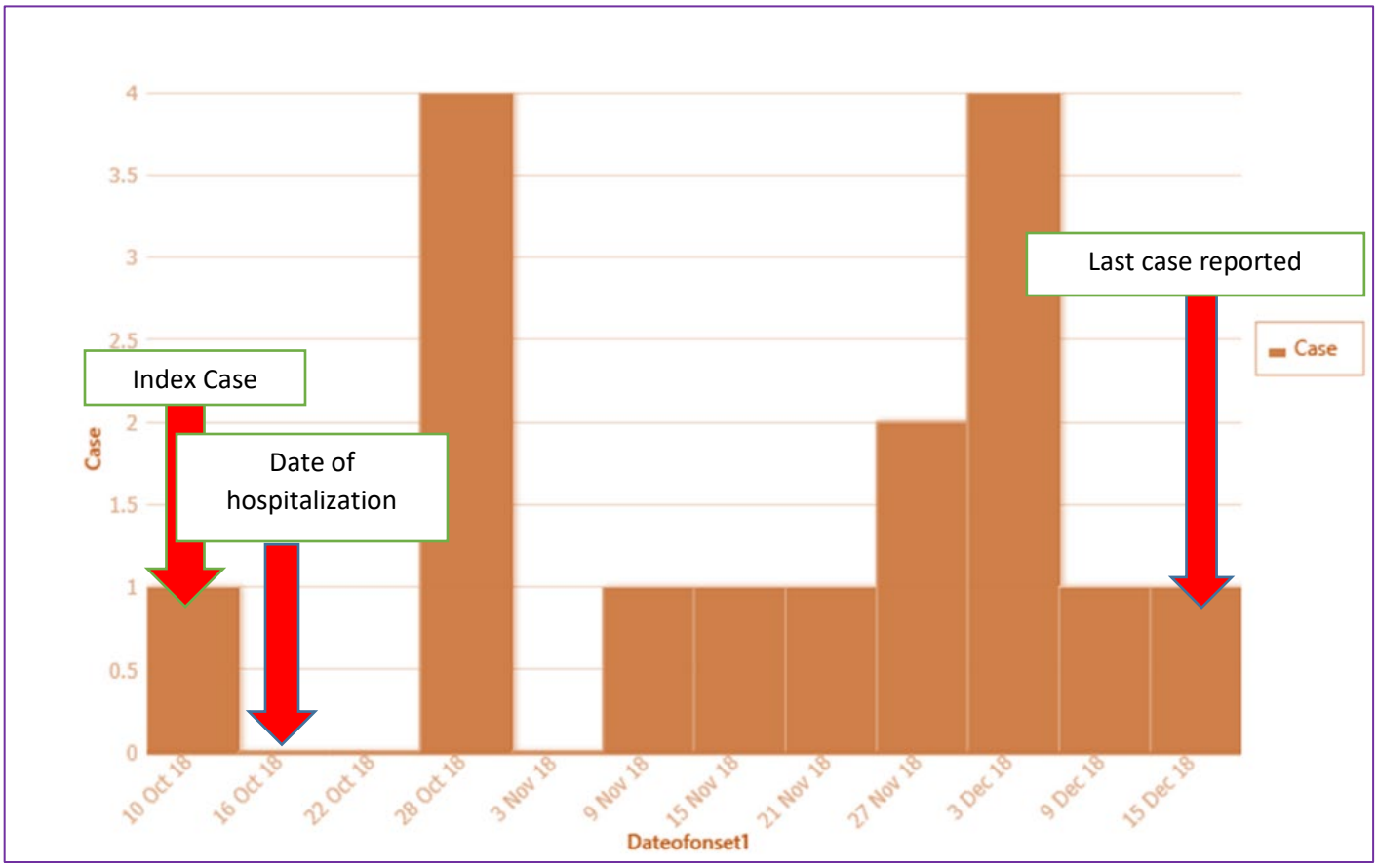

Figure 3: Epidemic Curve of measles outbreak, date of ones, hospitalization and last date of report, Mieso district, West Hararge zone, Oromia region, 2018

3 days seen the sign and Admitted to Asebot Hospital and followed by other cases with different age, from suspected case serum sample were collected and sent to EPHI and confirmed as positive to measles.

Fifty six percent of the case were and 32(67) of the control were female. Majority of the control 29(60%) were above 11 years old, Eighty seven percent of the case 14(87.5%) were under five years of age , only twelve percent of the case were between six and ten years old 2(12.5%). None of the case were married, only twenty one percent were married from control 21(43.75%). Only thirteen percent 2(12.5% of the case were attended the primary school, majority of the control were attended primary school and only twenty nine percent were does not attend any educational level.

Risk factor associated distribution showed, only 4(25%) of the cases had been travel history, fourteen percent 19(40%) of the control had travel history, Fifty nine percent of the case 14(87.5%) were not vaccinated against measles, Twelve point five percent 2(12.5%) of the case vaccinated only once. Only Thirty three percent 16(33%) of the control were not vaccinated, sixty seven percent 32(67%) of them were vaccinated.

Eighty one percent 13(81%) of the Case were with Sevier malnourished while only nineteen percent 3(19%) of them with no malnourished status. Only Eight percent 4(8%) of the control was with mal-nutritional status, while Ninety two percent 44(92%) of the control were with good nutritional condition. Seventy five percent 12(75%) of the case has no travel history, while sixty percent 32(67%) of the control had Travel history, ninety percent 3(19%) of the case of the case and eighty three percent 40(83%) of the Control had no contact with measles case. Sixty two percent 10(82%) of the case and eighty three percent 40(83%) of the control had no enough windows for ventilation of the living house.

Table 2: Child Mid Upper ARM Circumference (MUAC) Reading result of measles Cases in Mieso District West Hararge zone, Oromia region, 2018.

ID	Sex	Age(Yr.)	Reading (mm)	Interpretation
1	F	8	280	Normal
2	M	3	104	Sever
3	F	2	108	Sever
4	F	1	92	Sever
5	F	6 months	98	Sever
6	M	5	104	Sever
7	M	4	102	Sever
8	F	1	106	Sever
9	F	5	260	Normal
10	M	1	98	Sever
11	M	7	230	Normal
12	F	1	108	Sever
13	M	2	104	Sever
14	F	1	108	Sever
15	M	6 month	100	Sever
16	F	2	102	Sever

Table 3: Sociodemographic characteristics of measles case and control (Univariate Analysis) Mieso district West Hararge zone Oromia region, 10, October to 15, December, 2018.

Variable	Cases(n=16)	Control(n=48)
	Number (%)	Number (%)
Sex		
Male	7(44)	16(33.3)
Female	9(56)	32(66.7)
Age Group		
<1 yr.	2(12.5)	6(12.5)
1-5yr	12(75)	8(16.7)
6-10yr	2(12)	7(14.6)
11-15 yr.	0	4(8.3)
16-20 yr.	0	3(6.25)
21-25 yr.	0	8(16.7)
26-30 yr.	0	7(14.6)
>30 yr.	0	5(14.4)
Marital status		
Single	16(100)	27(56.25)
Married	0	21(43.75)
Widowed	0	0
Divorced	0	0
Educational level		
None	14(87.5)	14(29.2)
Primary 1-8	2(12.5)	18(37.5)
Secondary 8-12	0	16(33.3)

Table 4: Risk Factors Distribution among Measles Cases and Controls, (Univariate Analysis), Mieso District West Harage Zone Oromia Regional State 10, October to 15, December, 2018.

Variables	Case (N=16)	Control (N=48)
	Number (%)	Number (%)
Travel history 5 days before onset of the cases		
Yes	4(25%)	19(40 %)
No	12(75%)	29(60%)
They have received measles vaccination		
None	14(87.5%)	16(33%)
Once vaccinated	2(12.5)%	32(67%)
Nutritional status		
Sevier mal nourished	13(81%)	4(8)
Not mal-nourished	3(19%)	44(92%)
Exposure to measles patient with in a week before the onset of the case		
Yes	3(19%)	16(33%
No	13(81%)	32(76%)
The Living house has windows for enough ventilation		
Yes	6(38%)	8(17%)
No	10(62%)	40(83%)

1.1.4.3. LABORATORY INVESTIGATION

Out of totally 16 suspected measles cases, 4 serum sample were taken on 12, October, 1st and 11th of November, 2018, were sent to Ethiopian Public health institute (EPHI), and confirmed as positive to IGM (antibody) in laboratory.

1.1.4.4. Analytical Epidemiology (Case-control study)

We used all of the 16 cases (Median age: 2 years; IQR: 2; range: 0.6- 8) and recruited 48 controls (Median age: 15 years; IQR: 15; range: 0.6-42) for the case-control study. None of the demographic characteristics was associated with measles outbreak. Our multivariate logistic regression showed that the odds of developing measles were 3.917 times higher among those who did not vaccinated (Adjusted Odds Ratio**(AOR): 3.297 95% CI (0.97-0.989)**, and those with malnourished who develop measles case were **3.853** time higher than that of good nutritional status, (Adjusted Odds Ratio **(AOR):3.854, 95% CI(2.115-63.84)**

Table 5: Comparing the Distribution of Measles Risk Factors among Cases and Controls Using Bivariate and Multivariate Analysis, Mieso District, West Hararge Zone, Oromia Regional State, October-December, 2018

Variable		Case n (%)	Control n (%)	COR	P-value	AOR at 95% CI
Vaccination status	Yes	2(12.5)	16(33)	**7.75***	**0.005**	**3.917(0.97-0.989)***
	No	14(87.5)	32(67)			
Travel history	Yes	4(25)	19(40)	1.086	0.297	0.69(0.218-7.346)
	No	12(75)	29(60)			
Ventilation of the house	Yes	6(38)	8(17)	1.186	0.27	0(0)
	No	10(62)	40(83)			
Contact with Measles case within 5 days before onset of the case	Yes	3(19)	16(33)	2.897	0.089	0(0)
	No	13(81)	32(67)			
Nutritional status(Malnourished)	Yes	13(81)	4(8)	**12.157***	0.000	**3.853(2.115-63.84)***
	No	3(19)	44(92)			

Intervention of the Case:

All confirmation and clinically suspected measles case identified, the team from West Hararge Zonal and District PHEM officers , health workers intervene all cases were treated in Asebot Hospital, those with Sevier malnourished was supplemented balanced diet in the hospital.

Oromia health bureau was sent additional Vaccine dosage against measles and Mop up vaccine mass campaign were conducted especially child with under 10 years old for four affected Kebeles. As a result of intervention there was no any case reported for consecutive 30 days, the team declare that the outbreak ended in December 15, 2018.

1.1.5. **Discussion**

During Our outbreak investigation we identified the potential risk factor which may contribute for the occurrence outbreak, there is a malnutrition with in the kebeles since the community has less potential agricultural production to feed their family, dependent on aid, lack of balanced and surplus food plays a vital role for the repeated occurrence of the measles with in the district and current kebeles.

In addition to mal-nutritional factor there is shortage of vaccine supply, as a district level our team visited vaccine storage and handling condition and identified that there was shortage of Measles vaccine that was not enough to cover the whole children left unvaccinated. The team also checked the vaccine handing condition and storage that may influence the potency of the vaccine, In health posts where vaccine was conducted we have seen refrigerator in the health post with appropriate supply of power source from solar system and read optimal temperature.

From environmental and housing condition, there is no latrine in area community practiced OD near and around the residential area, living house was so close each other and confined with only few of the houses had Windows for ventilation having over crowded with extended family size average 6-10 family in a single house.

There is no any death from the total cases of an outbreak which indicates that lower rate than expected case fatality rate from developing country (3.6%) similarly there is lower attack rate which is expected to developing country. Based on the epidemiology of measles in Ethiopia and burden of disease modeling, it is estimated that more than 1.5 million cases of measles (all age)

and 70,000 deaths (assuming 4% case fatality ratio) would occur in Ethiopia annually. For many years the average number of measles cases reported to the Ministry of Health by the region ranged from 500-2000 annually (7)

According to the past study of measles. In Ethiopia, a seasonal pattern of occurrence of measles has been observed over the years, with increased number of measles cases during the late-early part of the year (December to February). Due to the low sub national routine measles coverage and prevailing poor living and nutritional conditions, measles outbreaks continue to occur frequently in different parts of the country, most especially in Oromia and SNNPR (southern) Regions where the density of the population is relatively high . The number of confirmed measles cases steadily increased from 73 in 2003 to a peak of 3,511 in 2008, following which there was a decline to 1,944 cases in 2009. Even though measles incidence rate has showed significant increase in the country since 2010 a dramatic increase is observed from 2012 up to 2014(8)

1.1.6. Conclusion and recommendation

From epidemiological point of view and past history of the area for the occurrence of the measles outbreak had been occurred repeatedly as a result of low coverage of vaccination and poor nutritional status were the potential risk factor for reoccurrence of the disease.

Based on the result and through investigation of the area we forward the following recommendations:

- Mieso district health office should improve the immunization coverage for all child, and MOP UP mass vaccination campaign for the rest of the children to control re-occurrence of the case with appropriate time and doses according to the standard and guideline of immunization.
- There should be separate balanced and supplementary diet especially for children under five years of age.
- Mieso District agricultural office should plan to improve crop production within the community.

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1.2. Anthrax Outbreak on Human and Animal in Farta and Smada Districts of South Gondor Zone of Amhara Regional State from March 10 to April 10, 2019

Abstract

Background: Anthrax is a serious zoonotic disease that can affect most mammals and several species of birds, but is particularly important in herbivores. A disease caused by the bacterium *Bacillus anthracis.* It is a gram-positive endospore forming, rod-shaped, non- motile, bacterium that grows in nutrient media, typically sheep or horse blood, under aerobic and anaerobic conditions with an optimal temperature of 35-37 ^{0}. The main objective of the study was to describe the cases in place, person and time and to confirm under laboratory.

Material and Methods: the study period was from March 13 to April 8, 2019. Descriptive study design methods was used, samples were taken and an individual sample DNA extracted from Tissue with QIAamp® DNA extraction mini kit, amplification was done with Real time PCR for confirmatory test.

Result: All samples from animal taken from March 13-April 8,2019 from Farta and Smada districts from two Kebeles were positive with Real time PCR, there are 17 cases from two districts 5 female and 12 male and one death of female of 45 year old were investigated. An index case was a person with 55 years old on March 13, 2019 duration of an outbreak is till 8 April, 2019. Conclusion and recommendation: All cases were confirmed of Anthrax, the two districts were an endemic area of anthrax, there was a death before two years with similar situation, since the vaccination coverage is under standard almost all livestock with in the outbreak and adjacent and human is still under risk. Awareness creation, routine vaccination of an animal is mandatory.

Key words: Anthrax, Smada and Farta district, descriptive epidemiology

1.2.1. Background

Anthrax is a serious zoonotic disease that can affect most mammals and several species of birds, but is particularly important in herbivores. The word anthrax is derived from a Greek word meaning charcoal or carbuncle. Anthrax, the disease, likely originated 6,000 to 7,000 years ago in Mesopotamia and Egypt, where agricultural civilization was first recorded. Scientific Literature first contained a reference to anthrax in 1769, by Fournier. Common names include: Malignant Pustule, Malignant Edema, wool sorters' disease, Rgpickers' Disease, and Maldi carbon.

Anthrax is a disease caused by the bacterium *Bacillus anthracis.* It is a gram-positive endospore forming , rod-shaped , non- motile , bacterium that grows in nutrient media, typically sheep or horse blood, under aerobic and anaerobic conditions with an optimal temperature of 35-37 ^{0} [1, 2]

Anthrax is a worldwide zoonosis that primarily affects herbivorous animals [3] in domestic animals there is high fatality in cattle, sheep goats, horses, donkeys and pigs [4] for wild animals. Omnivorous and carnivorous are moderately resistant although wild carnivore that scavenge from infected carcasses still succumb to illness [2]

Humans are known as accidental hosts and usual become infected by accidental contact with spores or vegetative bacilli either through the soil, insect or through direct contact with infected animal carcasses or material. Human at the highest risk of occupational exposure are veterinarian, livestock farmers, any personnel that handle animal product (such as butcher, wool sorter, tannery workers, etc.), and laboratory personnel [2]

Spores survive for decades in soil that is rich in calcium, has a PH greater than 6.0 and when temperatures are higher 15.5 ^{o} [5], it is considered a seasonal disease as outbreak tend to follow a prolonged hot and dry period that is followed by heavy rainfall. It is thought that heavy rain can carry spores during runoff in clumps of organic matter to concentrate in standing pools or puddles [5].

1.2.1.1. Transmission in Animals

B. anthracis naturally occurs in the soil, but its lifecycle almost exclusively takes place within the mammalian host [6], under the right conditions, spores can survive for years in the environment. Animal hosts acquire the disease through grazing, usually by ingestion or inhalation [6 and 7]. Once in a host, the spore are able to germinate back in to vegetative bacilli. The bacilli are then able to disseminate through the host system using toxin and capsule production. One the infection becomes systemic, the host dies from shock and hemorrhages blood and other bodily liquids. After death, the vegetative cells are exposed to air causing sporulation to occur and the spores return to soil for the next host [1 and 3]

In public health terms, the importance of anthrax lies in its ability affect large number of livestock and be spread rapidly across great distance [4] . Animal meat, hides, hair wool or bones may be transported long distance and spore can be carried with wind currents to new areas, especially during dry periods [4]

There are two main routes of transmission in animals: ingestion and inhalation. It is generally accepted that animals acquire anthrax through contact with spores in the soil and that some sort of lesion is necessary for establishment of an infection, as *B. anthracis* is considered non-invasive [8]. Insects are also play a role in transmission, primarily through blow flies. Animals that have died or are dying from anthrax provide the main source of infection for animal that through shedding of bacilli, which eventually becomes spore, in to the environment.

1.2.1.2. Transmission in Humans

There are three major routes of transmission for humans: ingestion, inhalation and cutaneous. The most common form of disease is cutaneous which occurs for about 95% of human cases [3 and 7] This form is acquired through abrasion or open wounds on the skin that come in to contact with either vegetative cells or spores. Cutaneous anthrax has a mortality rate of 5-20% for untreated case and <1% in treated case [7] while rare, cutaneous anthrax can produce shock.

Ingestion anthrax develops after ingestion of spores or cells through contaminated meat. While uncommon, the mortality rate is through to be much higher than cutaneous at 25-60 % [7].The most fatal type of infection with anthrax is through inhalation which occurs from breathing in

spores from the environment. Natural infection through this route is rare but intentional release has occurred which lends to a greater risk of large scale outbreaks. The mortality rate for inhalation anthrax varies, with estimate of about 45% with antibiotic treatment and as high as 97% without.[9]

Aside from the major routes of transmission there are also several less common routes of infection for humans. Insertional anthrax is becoming more common among IV drug users which characterized by subcutaneous infection of soft tissue, this route presents differently than cutaneous infection, and has shown to be harder to treat , with infectious leading septic shock, meningitis and death in 34% of patient despite treatment [7]

Clinical progression in Animals

In Animals the most common route of infection is inhalation, the incubation period can range from as little as 36-72 hours following the entry of the bacteria or spore to 1-14 days, the typical incubation period in livestock is 3-7days. Once in the blood stream, cells can have a doubling times as fast as 0.75-2 hours depending on the host [1]

Clinical presentation and susceptibility depends on the animals species affected. Cattle and sheep are highly susceptible and can have hyper acute to acute disease including sudden death without prior signs of illness. Horses have hyper acute to acute diseases with sign for 2-3 days before death.

Signs of acute infection in domestic animals include distress, loss of appetite, disorientation, difficulty in breathing, reduce milk production, subcutaneous hemorrhage, and swelling in the submandibular fossa with or without rise in body temperature .[8]

Clinical progression in Humans

Cutaneous anthrax presents in as little as 12 hours to as long as 19 days after initial infection, but is typically 1-12 days, a small painless skin lesion with swelling usually appears on exposed regions of the body such as the face, neck, arm or hands . [10]

The lesion eventually dried and forms a black center, called an eschar which sloughs in 2-3 weeks, during this periods fever can occur but is rare, in small percentage of cases, septicemia can develop. It should be noted that toxin can be detected in the blood stream even when systemic disease is not present.[11]

Ingestion anthrax has an incubation period of about 2-5 days, there have been documented outbreaks where the incubation time is as little as 42 hours. [7]it can be presented in two different ways: intestinal and oropharyngeal. During intestinal anthrax, vegetative cells cause ulcers or lesions throughout the small intestine, symptoms include nausea, vomiting, anorexia and fever, in severe cases abdominal pain, hematemesis, bloody diarrhea and septicemia can present. Oropharyngeal anthrax occurs when vegetative cells settle in pharyngeal area and produce ulcers, patients usually with fever, neck swell]ng and sore throats.[10]

Inhalation anthrax (also known as pulmonary) has the highest mortality and has historically been linked to industrial cases of diseases, although bioterrorism has become the greatest concern in large scale outbreaks.[7 and 9] Inhalation cases take a biphasic clinical course, after approximately four days, patients develop flue like symptoms with fever, cough and myalgia, and mediastinal widening is commonly seen during this phase. Inhalational anthrax can be successfully treated if antibiotics are started in this stage of illness but diagnosis is difficult.

Recently a new type of anthrax infection called injectable anthrax has been documented and is associated with IV drug users, this infection seems to be different than cutaneous diseases with increased risk of shock and higher mortality rate despite antibiotic treatment. [7] surgical intervention is a common treatment, which shows prominent tissue edema, diffuse capillary bleeding and necrosis of superficial adipose tissue.

1.2.2. Objective

General Objectives:

- To investigate anthrax outbreak and to confirm suspected cases under laboratory from animal and human and propose possible control measure.

Specific Objectives:

- To investigate anthrax outbreak from human and animal cases and describe in place, person and time
- To detect and confirm in laboratory for the first time in Ethiopian condition.
- To give awareness and recommendation for the community who were living in anthrax endemic areas

1.2.3. Material and Methods

1.2.3.1. Study area and period

The Outbreak was occurred in Farta and Smada Districts of Wowa Magera and Dudubia kebeles of south Gondor zone Amhara regional state with in the rural community from March 25-April 7, 2019. Farta district located about 20 km distant from Debre-tabor to the south part, Smada district located with same direction which distant about 60 km out of which 40 km all weather road and 20 km no access to road with step and ragged geographical feature.

1.2.3.2. Study design; descriptive cross-sectional study design was conducted

1.2.3.3. Data collection Methods

Structured questioner and interview was made with people from Wowa Magera and Dudubia Kebele who were living in the vicinity of the case and exposed to the case and district and Kebele veterinarian and Paravet about the vaccination status for animal and health post and extension workers in human cases to collect basic epidemiological data, date of onset, clinical feature, data collection was made in pre designed format to collect, demographic data, livestock population, vaccination history of animal, onset of the disease and hospital visit of inpatients detail history.

1.2.3.4. Laboratory diagnostic method:

1.2.3.4.1. Sample collection: Sample was collected from animal by veterinarian from Goat and cow dead of suspected anthrax, and Human sample swab collected in patient who admitted to Debre-tabor Hospital both human and animal samples were kept in cold chain and transported to Ethiopian Public Health Institute (EPHI) and National Animal health Diagnostic and Investigation center (NAHDIC) in Biohazard bag with triple package.

1.2.3.4.2. DNA Extraction: DNA Extraction was performed from Tissue with appropriate biosafety measure and decontamination with 10% bleach in BSL-3 zoonotic laboratory, each sample were individually extract with QIAamp DNA Extraction mini kit. Tissue was rinsed with PBS to avoid contamination of environment and lab personnel inhalation of spore from dried tissue, then minced with sterile scissors and grinded with mortar and pistil with fine sterile sand, after formed suspension, Extraction started with lysis buffer added 180 micro liter of ATL buffer,

then 20 µl proteinase K and vortexed. Decontaminate with 10% bleach then incubated with 56°c for 1-3 hours vortex every 30 minutes.

After 3 hour incubation in water bath at $56^{o}_{c,}$ the whole volume was filtered with Millipore to avoid spore not to be extracted with other cells, transfer to new mini-spin column added 200 µl of Al buffer, vortex then for binding added 200 µl of absolute alcohol (Ethanol) vortex centrifuge 8000rpm for 3 minutes. After binding step followed by washing one (AW1) centrifuge at 8000rpm for 3 minutes, Aw2 with similar procedure. Finally elusion was done with 200 µl of AE buffer followed by centrifuge at 8000rpm for 3 minutes and keep DNA at -20^{0}_{c} till next step.

1.2.3.4.3. DNA Amplification

Bacterial amplification was performed with Real-time PCR comprised with Master-mix components; primers which targeted for both protective antigen (PA) and Capsular antigen (CAP) both forward and reverse primers and probes for each.

Protective antigen (PA)

PA- Forward 5 ' CGG ATC AAG TAT ATG GGA ATA TAG CAA - 3'

PA- Reverse 3' CCG GGT TAG TCG TTT CTA ATG GAT 5'

PA-Probe: FAM CTC GAA CTG GAG AGT GTT ACC GCA AAT-BHQ1

Capsular antigen (CAP)

Cap-Forward: ACG TAT GGT GTT TCA AGA TTC ATG

Cap-Reverse: ATT TTC GTC TCA TTC TAC CTC ACC

Cap Probe: FAM- CCA CGG AAT ATC TCA AAT GGC AT-BHQ1

In Master-mix component 10X Exo IPC Mix and 50X Exo IPC Mix DNA were included to monitor inhibitory substance with in Real time PCR. Thermal cycler protocol was Denaturation 95°c for 10 minutes, Amplification 95°c for 15 seconds and 60°c for 1 minutes for 40 cycle's standard mode of 7500 with passive reference of ROX. Volume per run 20µl of Master-Mix components 5µl of unknown sample with total volume of 25µl.

1.2.4. Case definition:

According to Guideline for surveillance and outbreak management of Anthrax in human and animals in Ethiopia

Clinical and post-mortem anthrax case definitions

A case of sudden death of animals accompanied by bleeding (un-clotted dark tarry blood) from natural orifices, absence of rigor mortis, and rapid bloating of carcasses should be regarded as suspected anthrax cases.

Confirmed Anthrax case definition

A confirmed case of Anthrax requires detection of Bacillus anthracis either in polychrome methylene blue stained blood smear supported where possible with molecular confirmation by polymerase chain reaction (PCR).

Standard Case definition according to PHEM/WHO

Suspected case: Any person with acute onset illness characterized by several clinical forms which are:

Localized form:

Cutaneous Anthrax: skin lesion evolving over 1 to 6 days from a papular through a vesicular stage, to a depressed black eschar invariably accompanied by oedema that may be mild to extensive

Systemic forms:

Gastro-intestinal Anthrax: abdominal distress characterized by nausea, vomiting, anorexia and followed by fever.

Pulmonary (inhalation) Anthrax: brief prodromal resembling acute viral respiratory illness, followed by rapid onset of hypoxia, dyspnoea and high temperature, with x-ray evidence of mediastinal widening

Meningeal: acute onset of high fever possibly with convulsions, loss of consciousness, meningeal signs and symptoms; commonly noted in all systemic infections

And has an epidemiological link to confirmed or suspected animal cases or contaminated animal products.

Confirmed case:

A suspected case that is laboratory confirmed with Culture or ELISA or Western blot, toxin detection, chromatography assay, fluorescent antibody test.

Community case definition:

A person who gets ill within 7 days after eating meat of sick animals or close contact with animals that have bleeding from nose, mouth and anus.

1.2.3.4. Data Processing and Analysis: The data were entered and analyzed using Epi-Info7 ™ version 7.1.0.6. Results were presented using graph and table. Attack rate and case fatality rate were also calculated.

1.2.5. Result

Demography

The total population of an area where an outbreak occurred in Farta district Wowa Magera 7336 of which 4658 (63.5%)Female and 2003 female in Smada District of Dudubia PA there are about 4009 out of which 2006(50%) is male. With regards to population of livestock there are 13,221, in Wowa Magera and 30,976 livestock in different species as indicated in Figure 4.

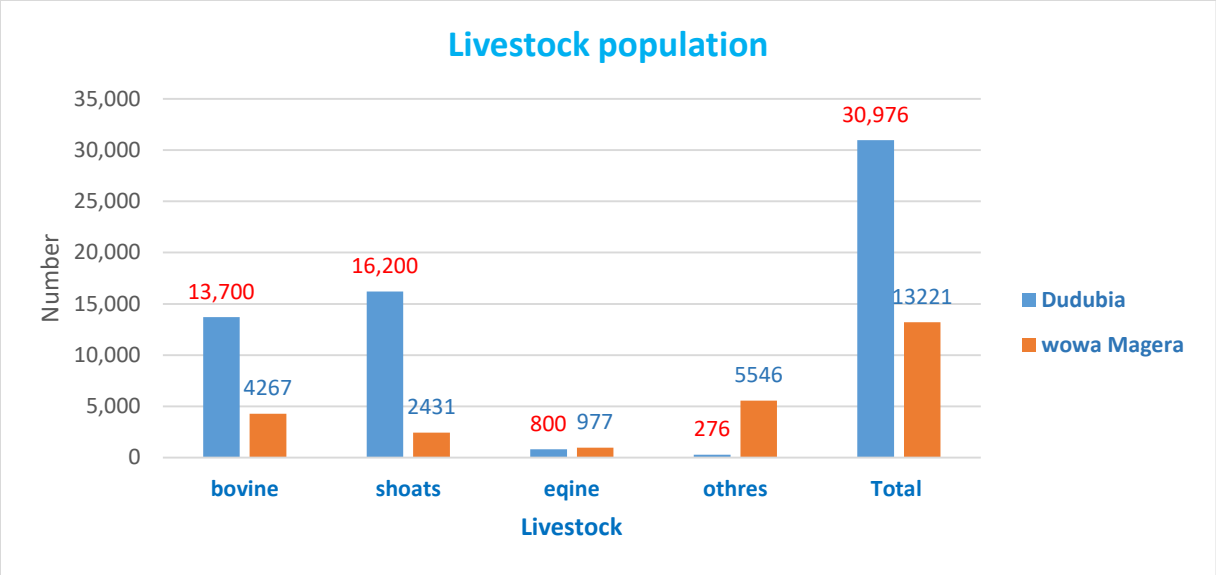

Figure 4: Livestock Population under Risk of Anthrax in South Gondor, Amhara Region 2019

From the two district, two kebeles and adjacent kebeles those who were close proximity with Geographical and social interaction with the area where an outbreak happened were 6 Kebeles totally 18,216 people were under risk area who prone to be exposed for anthrax spore directly or indirectly. As Indicated in Figure 5.

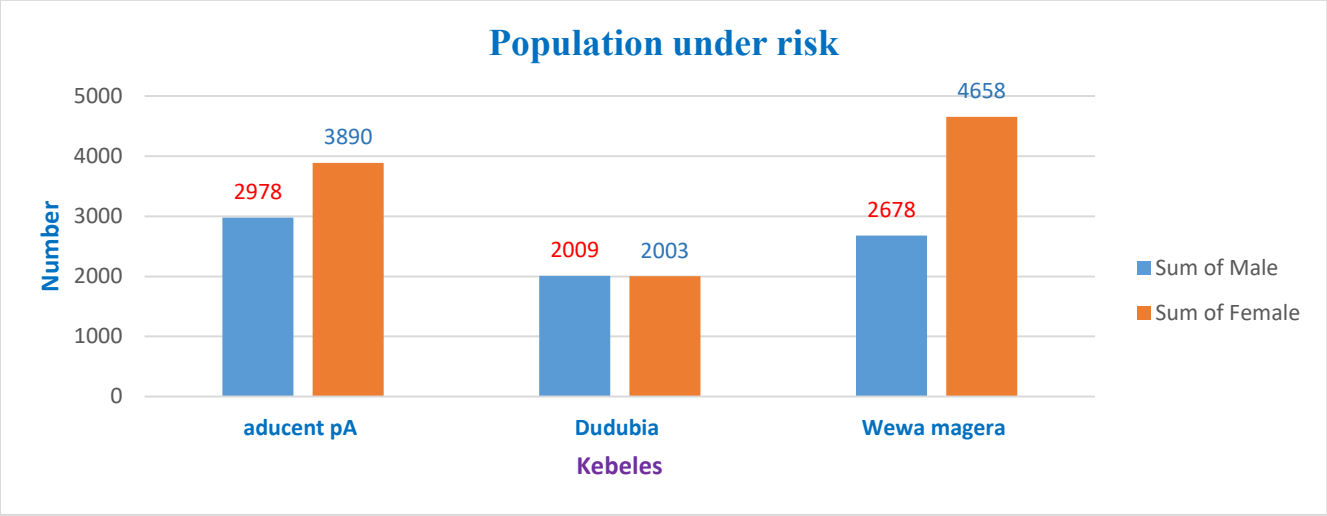

Figure 5: Population under Risk of Anthrax South Gondor, Amhara Region, 2019.

Vaccination coverage for livestock with in the Kebele where an outbreak occurred who were vaccinated with in six month before the onset of an outbreak, cattle 23.4% shoats 49.85%, and none of equine were vaccinated.

1.2.5.1. Descriptive by place:

Anthrax case were detected from Farta and Smada districts of south Gondor, in two kebeles after contact of animal dead of one goat and one cow. An anthrax case from two districts the higher case in people who were in Farta district of Wowa Magera Kebele.

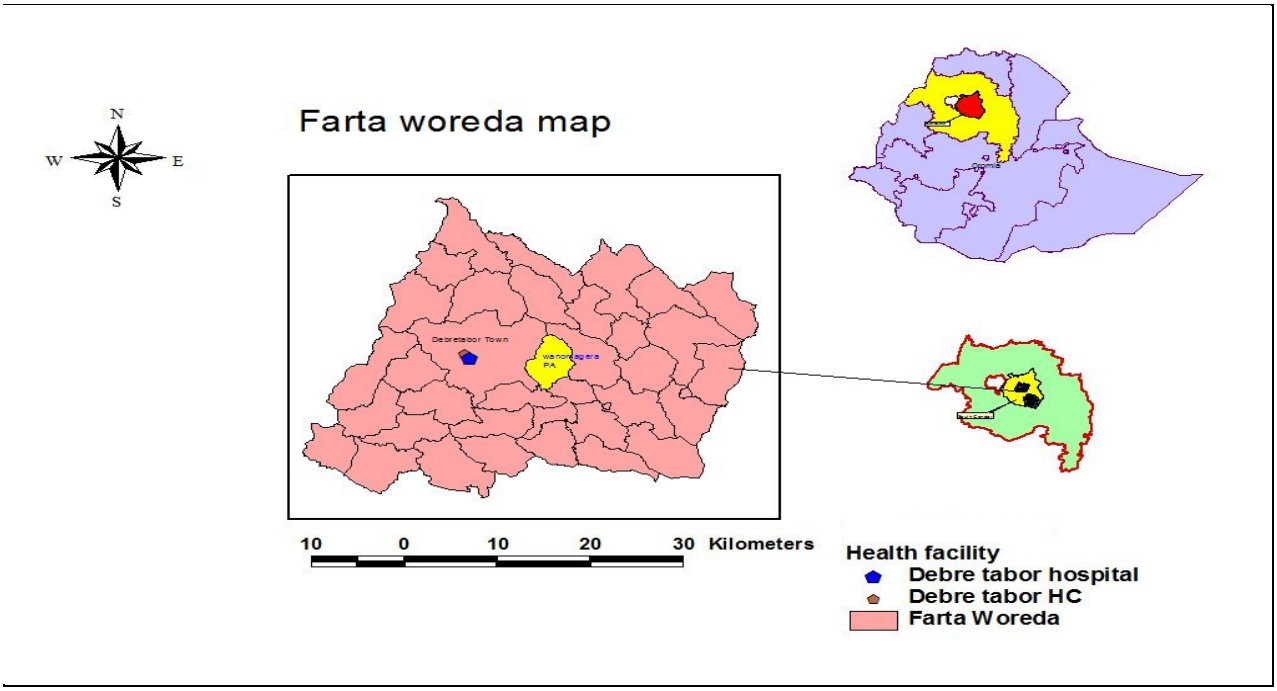

Figure 6: Wowa Magera Kebele, Farta District, South Gondor zone Amhara region, April, 2019.

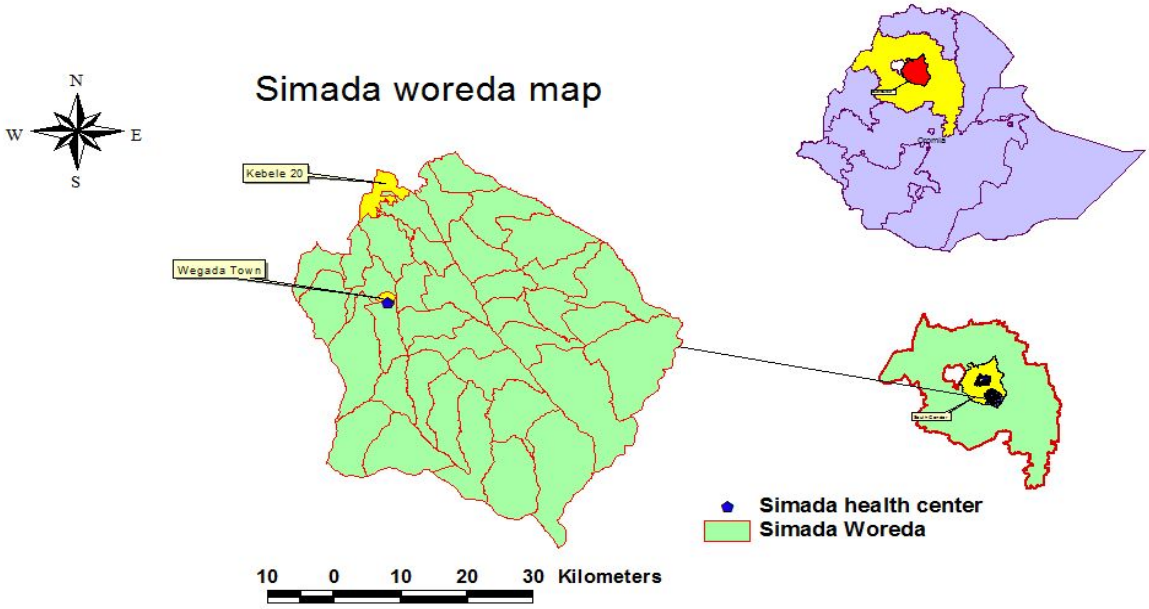

Figure 7: Map of Dudubia Kebele, Smada District, South Gondor, Amhara region April, 2019.

1.2.5.2. Descriptive by person:

There were 18 individuals who develop cutaneous anthrax out of which 17 from Farta district and 1 from Smada districts after contacted with sudden death cow from Farta and Goat from Smada district of Dudubia Kebele. Out of the total case who had exposed, female around 42 years old who did not visit any health facility were die due to anthrax.

Table 6: Anthrax Case Of Wowa Magera Kebele, Farta District, South Gondor Zone, Amhara Regional State, April, 2019

Wowa Magera Kebele	**Population**	**Case(n)**	**Percentage**	**AR/1000 Population**	**Death**	**CFR**
Male	2678	12	70.59	4.5	0	0
Female	4658	5	29.41	1.1	1	20
Total	7336	17	100	2.32	1	5.9

Table 7: Anthrax Case of Dudubia Kebele, Smada District, South Gondor, Amhara Region, April, 2019.

Dudubia Kebele	**Population**	**Case**	**Percentage**	**AR/1000 population**	**Death**	**CFR**
Male	2003	1	100	0.5	0	0
Female	2006	0	0	0	0	0
Total	4009	1	100	0.25	0	0

Since all case was cutaneous type of anthrax the picture taken from patient who was admitted in Debre Tabor hospital of 55 years old male having Chancker scar on his left hand as attached picture 1 and a child who aged 7 years female sign on eye illustrated on Figure 7.

Figure 8: Patient with cutaneous anthrax in patient taken from Debre-tabor Hospital April 7, 2019.

Cutaneous anthrax based on our questioner survey sign and symptom of the onset of the disease 95% of them develop swelling with CI of 95% as a result of statistical data showed in Table 3.3. And 88.24 % seen itching with Level of Confidence 95% as indicated in table 7.

Table 8: Frequency of Respondent's Anthrax Case Swelling Sign, Wowa and Dudubia Kebele of South Gondor, 2019.

Swelling	Frequency	Percent	Cum. Percent	Exact 95% LCL	Exact 95% UCL
Yes	16	94.12%	94.12%	71.31%	99.85%
No	1	5.88%	100.00%	0.15%	28.69%
TOTAL	17	100.00%	100.00%		

Table 9: Frequency Of Respondent's Anthrax Case Itching Sign, Wowa Magera Kebele Farta District, South Gondor Zone, Amhara Region, 2019

Itching	Frequency	Percent	Cum. Percent	Exact 95% LCL	Exact 95% UCL
Yes	15	88.24%	88.24%	63.56%	98.54%
No	2	11.76%	100.00%	1.46%	36.44%
TOTAL	17	100.00%	100.00%		

1.2.5.3 Descriptive by Time

The onset of human outbreak was 5 days post exposure of dead animal carcass which is a cow dead on 13 of March 2019, first case was on 19 of March,2019 three cases were visit to hospital followed by others with different date till 3rd of April 2019, as others discharged from hospital in average of 10-12 days of inpatient, one serious case from Dudubia of Smada district admitted on March 27 to April 8 still in hospital for 12 days not yet discharged till our last visit on 8th of April,2019.

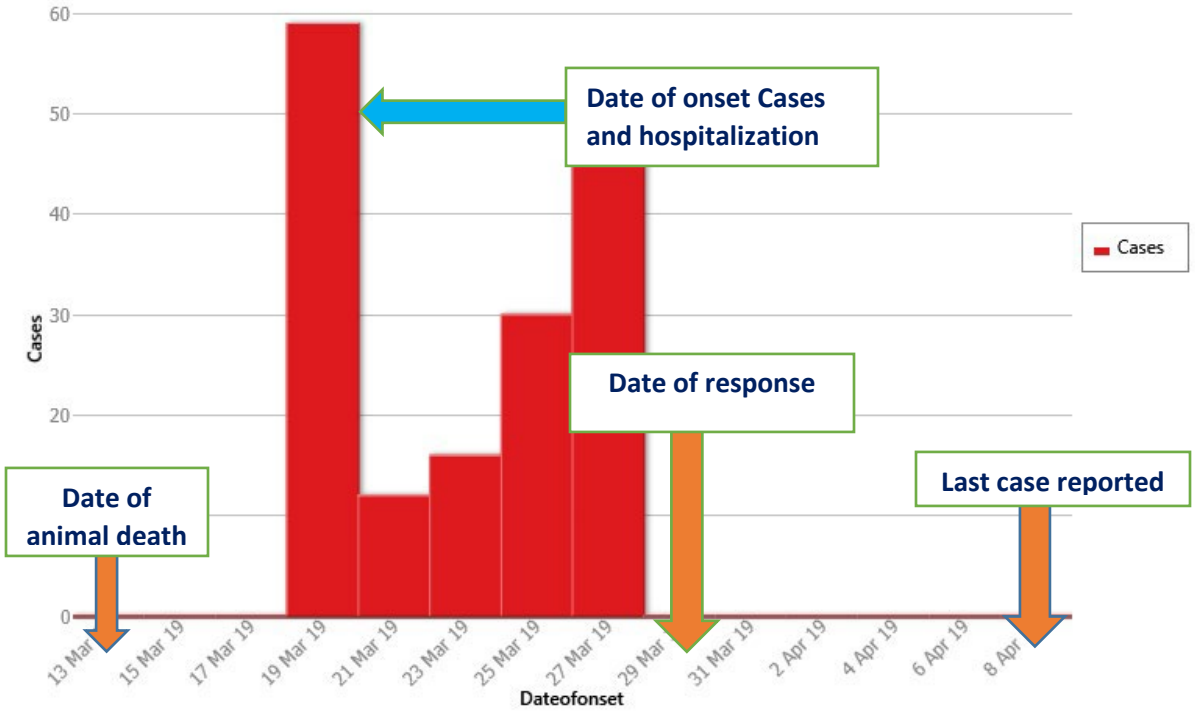

Figure 9: Epi Curve of Anthrax outbreak date of onset in Farta and Smada districts of South Gondor, Amhara Region, March-April, 2019.

1.2.5.4. Laboratory Result

From two Anthrax suspected animal Cases sampled and four human samples were examined and confirmed with RT-PCR, targeted regions of Protective antigen (PA) and capsular antigen (CAP). This was the first time that an anthrax were confirmed as a national wise with Real time PCR.

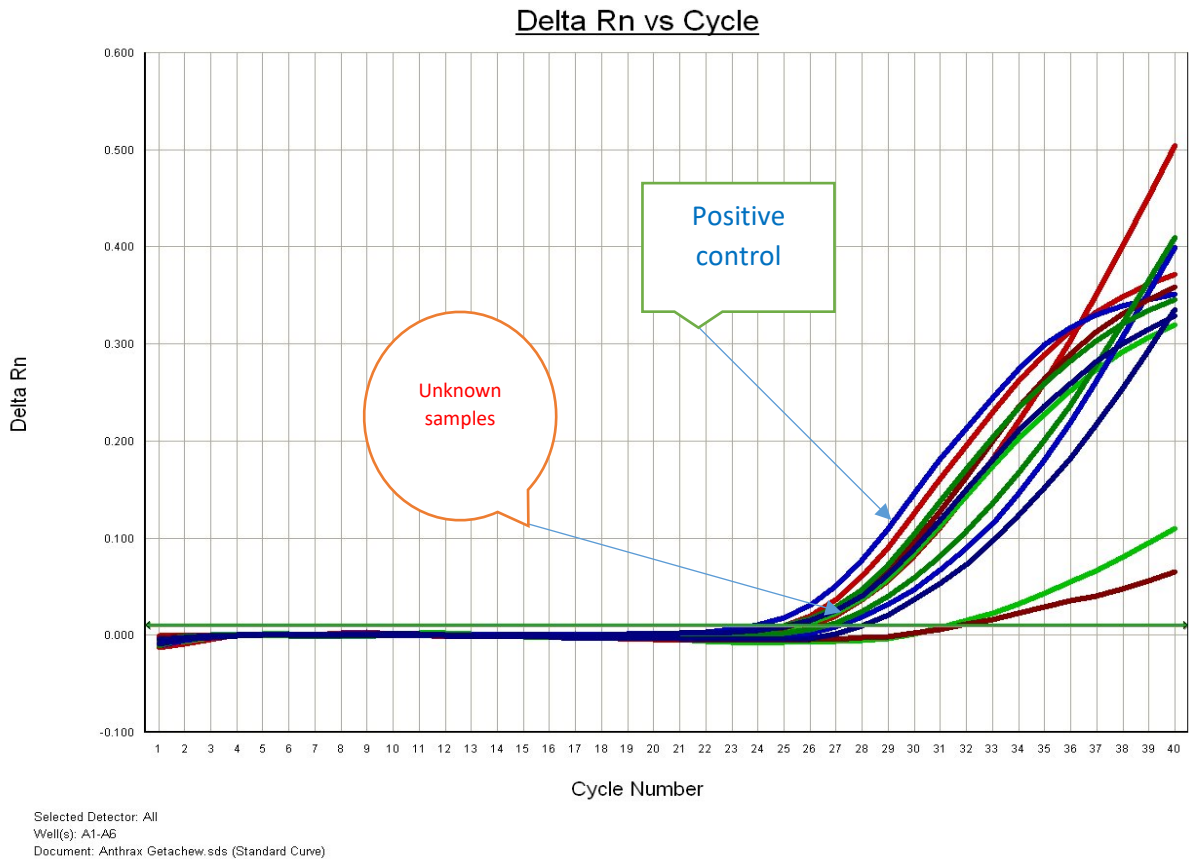

Figure 10: Anthrax laboratory result of RT-PCR DNA amplification plot.

Intervention and Control Measure:

In both Districts of anthrax case animal carcass were collected from all individual houses and burned and buried in deep well. Because this will be the potential source of animal outbreak, the community who is living in the area was very prone to be affected by Anthrax spore.

All animas with in the Kebeles and adjacent were under mass vaccination campaign since the source of the case was animal and an animal could get the source of infection from the grazing land and soil of contaminated area that viable for decades in the soil in spore form.

In both districts of the community, animal and human health workers, there was mass awareness creation and multidisciplinary approach was linked among different sectors with zonal, district and Kebele level.

1.2.6. Discussion

The Vaccine coverage of animal against Anthrax was under the acceptance range since majority of animal is not vaccinated this plays a great contribution for the outbreak, as epidemiological surveillance of the animal health indicated that the area is endemic to anthrax there was an outbreak two years ago and Couse for death two persons.

South Gondor zone is one of the area among traditional tannery was widely practiced out locally processing animal hide and skin for local commodities as a result of this most of the people who were contact with skin and hide had cutaneous form of anthrax endemically, since there is no routine report and lack of awareness human cases were under report with health facilities.

A century's experience of the natural disease in humans suggests that it probably does not take many spores to initiate a cutaneous infection once the necessary access to sub epidermal tissues is achieved, despite the implication otherwise in the experimental data. It is generally accepted that *B. anthracis* is non-invasive and that the spores must gain access to sub epidermal tissue through a cut or abrasion before infection can occur, although this has been challenged: in a series of laboratory-acquired cutaneous anthrax cases, preceding trauma at the site of the lesion was noted by only 5 of the 25 patients [12].

Anthrax eschars are generally seen on exposed unprotected regions of the body, mostly on the face, neck, hands and wrists. Generally cutaneous lesions are single, but sometimes two or more lesions are present. For example, with infection resulting from skinning an infected dead animal, multiple lesions may be seen on hands, wrists or arms. The incubation period ranges from as little as 9 hours to 3 weeks, mostly 2 to 6 or 7 days. In the October–November 2001 anthrax letter cases in the USA, the mean/median incubation period for the 11 cutaneous cases was 5–6 days, with a range of 1–12 days [13-15].

The cutaneous anthrax lesion commences as a minute red macule which then becomes a tiny papule or pimple, heralded by pruritus, often present before evidence of any visible lesion. These papules then enlarge to take on a pinkish fleshy appearance followed by formation of fine, tight, shiny yellow or pink vesicles. Then a non-pitting brawny edema begins to appear; and satellite vesicles or blisters may also appear. By about 48 hours the central vesicle ruptures forming a moist ulcer, with a reddish or brownish base and later the base of the central ulcer darkens and by the

fourth day, it is black and is surrounded by a raised erythematous indurated ring of tissue. At this stage the lesion is characteristic of anthrax [16]

Anthrax cannot directly transmit from human- to-human contact. Rather, it is transmitted by the transfer of spores from infected humans or animals to potential hosts or by exposure to airborne anthrax spore[17]

Spread of the organism with in an area may be accomplished by streams insects, dogs, other carnivores, wild birds and by fecal contamination from infected animals. Introduction of infection into a new area is usually through contaminated animal products such as bone meal, fertilizers and hides or by contaminated concentrates or forage (18)

Since there is no human vaccine in Ethiopia, and source of infection is animal after exposure of spores that prone to animal to be a case, the current outbreak shoed us an animal of sudden death was slaughtered and shared to the community, for human consumption since no one had consumed meat of dean animal all exposed patients developed cutaneous type of anthrax. All patients who visited hospital after suspects to anthrax recovered except one female who stay about 20 days post exposure and ill without visited and health facility, as case became worsen she decide to visit health facility but die of anthrax on the way to visit hospital, in some literature there is a case fatality rate of 5-20% if cutaneous anthrax left untreated.

Conclusion and Recommendation

This outbreak investigation was the one which confirm Anthrax for first time in Ethiopia with real time PCR test method. All cases were confirmed cases, with regards to the risk factor as South Gondor is Endemic area for anthrax, there is spore in the soil, dead animal hide and skin, feed and from the environment. As a vaccination coverage is the lowest status it play a vital role for the source of occurrence of the outbreak.

The team also identify that the community has no any awareness of anthrax, its severity, way of transmission, source of infection, availability of treatment in health facilities, with the current outbreak we recognized that one Female adult of exposed dead animal carcass followed traditional healer after onset of the disease, after several days of trial die of cutaneous anthrax. This indicates that almost all patient who visit hospital had recovery after inpatient of 10-11 days.

Based on our finding and assessment of the area the following points were forwarded to the District health office, community and administrative officials and development agents:

- An animal with sudden death with prognostic sign should not be opened, buried with intact skin and hide in deep with clay soil to avoid exposure of scavenger and drainage to rivers and streams.
- As an area and adjacent district were under risk an animal should regularly vaccinated every six month since source of infection is an animal.
- District PHEM and veterinary clinic should give routine awareness creation for the community for better control of the disease in each level.
- We recommend also for health facilities to think about the human vaccine for further prevention of the public health.

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CHAPTER 2. SURVEILLANCE DATA ANALYSIS

2.1. Surveillance data analyses of Malaria in Sebeta district from 2012-2017.

ABSTRACT

Back ground

Malaria is one of an endemic and sever disease in Ethiopia which covers about 75% of the total area and more than 54 million people are population under risk. The transmission is seasonal as the disease affects people of the productive age has a great impact in the economy as a result of illness people lack of labor stop in working, medical expense, school dropout from year to year.

Methods

Sebeta health center was selected and an individuals of the previously documented malaria cases were collected and data entry and analysis were performed in Excel and epi.info 7.1 software.

Result

Out of examined 35,469 patients during 2012-2017, 1947(63%) were male, 1143(37%) female were positive to plasmodium, out of confirmed positive cases 2140(67%) *vivax* and 1069(33%) were *falciparum*, malaria trend of the area indicated in year 2016 was the most significant followed by 2017 while the least prevalence showed in 2013.

Conclusion

The trend of the malaria cases is increasing from time to time both in species of *vivax* and *falciparum* in both genders. Since the altitude of the Sebeta area is not considered as low land the most prevalent is vivax this figure may indicate us some of the relapsing cases may re-examined and *falciparum* case was detected from people of immigrant from other malaria endemic area.

2.1. INTRODUCTION

Malaria is one of the most severe public health problems worldwide with 300 to 500 million cases and about one million deaths reported to date, 90% of which were reported from Sub Saharan African countries. It is the fourth leading cause of death of children under the age of five in developing countries [1]

It is a major concern the major diseases of poor people in developing countries and one of the leading causes of avoidable death, especially in children and pregnant women. Sub-Saharan Africa carries the bulk of the global malaria burden, 71% of cases and 86% of global deaths. A person in Africa dies of malaria every 10 seconds.[2]

It is estimated that three-fourths of the land below 2000 meters is malarias' with two-thirds of the country's population at risk.[3]. This makes malaria the number one health problem in Ethiopia with an average of 5 million cases a year [4] and 9.5 million cases per year between 2001and 2005. The disease causes 70,000 deaths each year and accountant for 17% of outpatient visits to health institutions. It also accounts for "…15% of admissions and 29% of inpatient deaths" a figure considered to be too low given that more than a third of the country's population does not have access to health services [5].

2.2. Objective

General Objectives:

The main objective of the study is to describe the trend and magnitude of malaria in Sebeta district from 2012 to 2017.

Specific Objectives:

- To describe the trend of Malaria cases in Sebeta district from 2012 to 2017.
- To determine the magnitude of the malaria cases in Sebeta district.

2.3. Literature Review

Malaria is caused by a protozoan belonging to the genus *Plasmodium*. Four species: *Plasmodium falciparum*, *P. vivax*, *P. ovale*, and *P. malariae* infect humans but they each differ in many aspects of their biology and geographic distribution. "*P. falciparum* is found in most tropical regions throughout the world, and is the most dangerous of the four in terms of both its lethality and morbidity." All undergo two forms of replication: sexual and asexual. The, parasites develop optimally in the vector but cease developing at temperatures 16°C or below. "High humidity prolongs the life of the vector and transmission is extended under these conditions. In the human intermediate host, the parasite must function at 37°C or higher, since the infection induces a significant rise in core temperature during the height of the infection."[6]

Malaria transmission intensity, along with its temporal and spatial distribution in Ethiopia, is mainly determined year ease by the diverse eco-climatic conditions. Climatic factors such as temperature, rainfall and humidity show high variability mainly as a function of altitude and are the most important variables that influence malaria transmission. [7].

In Ethiopia major epidemics occur every 5 - 8 years, but focal epidemics were occurring every year. *P. falciparum* and P. vivax are the two species commonly known to cause malaria in Ethiopia accounting 60% and 40% proportion, respectively [8]

The distribution and transmission of malaria varies from place to place. For example, the distribution of malaria in Ethiopia is largely determined by altitude. Altitude affects the pattern of malaria distribution in Ethiopia through its effect on temperature[9]

Malaria is a major concern in the country since it is one of the leading causes of morbidity and mortality. Despite the current efforts to control malaria in Ethiopia, the situation has not improved mainly due to the increasing problems of parasite resistance to the relatively cheaper anti-malarial drugs, vector resistance to insecticides, low coverage of malaria, poor access to health care, service infrastructure large and limited financial and human resources [10].

Mosquitoes that carry malaria are most active at night and in the early morning; bed nets are particularly vital during these times. Studies show that sleeping under a bed net can reduce child mortality from malaria by as much as 20%. The repellent in the nets can also reduce the number of mosquitoes in the surrounding area [11]. When 80% of households use bed nets in a community,

studies suggest that mortality from malaria for those living within 300 meters is significantly reduced. [12].

Malaria has been a perennial cause of human suffering and mortality for millennia. The Greeks were the first to write about it, and Egyptians hieroglyphs also made a mention of it. It exists throughout much of "…tropical and sub-tropical regions of Africa, Asia, and South and Central America." [13]

As is the case everywhere else where malaria is endemic, the disease is far from being conquered. The agent plasmodium has developed resistance to a number of drugs while the vector mosquito has learned to fend off the chemical onslaught launched by humans. The early 21st century fight in Ethiopia was guided by the Abuja (Nigeria) declaration with the following targets for the year 2005 [14]

The mosquito life cycle has four distinct developmental stages: egg, larva, pupa and adult are about 0.5 mm in length, boat-shaped and nearly all species are provided with tiny air-filled floats that allow them to remain on the water surface. Eggs are laid singly by the female *Anopheles* on the type of water preferred by a particular *An. arabiensis* usually prefers clean rainwater and open sunlit habitats without vegetation for oviposition.

The larvae hatch from the eggs as small and have a distinct head and thorax, and an abdomen composed of nine segments. The globular thorax is broader than the head or abdomen and somewhat flattened. It has several groups of hairs that are useful in identifying the species. Each abdominal segment has hairs, which are useful for distinguishing different species of *Anopheles*. The body of an anopheles larva lies parallel to the water like all mosquito larvae, those of *Anopheles* undergo three successive molts, separating the life of the larva into four stages or instars, i.e. first instar, second instar, third instar and fourth instar, which mainly differ from each other by the size of the larvae. At the end of the fourth stage, the larva changes into a pupa. The pupa is comma-shaped and differs greatly from the larva in appearance.

Pupae do not feed during their aquatic existence, but come to the water surface to breathe. Finally, the pupa emerges as adult stage. The length of each stage is dependent on a range of environmental conditions, including temperature. In favorable conditions, it takes an average of seven to ten days from egg to emerging adult.

2.4. METHODOLOGY:

2.4.1. STUDY AREA:

The study was conducted in Sebeta district, Oromia special zone or Oromia regional National state. An area was located in south West part of Addis Ababa which is 27 km distant from the capital city of the country. The district has four governmental health centers, Daleti, Alemgena, wolette and Sebeta. a data source of the positive confirmed case were those patients diagnosed in all health facilities in district governmental and private, which compiled in Sebeta town health center PHEM data.

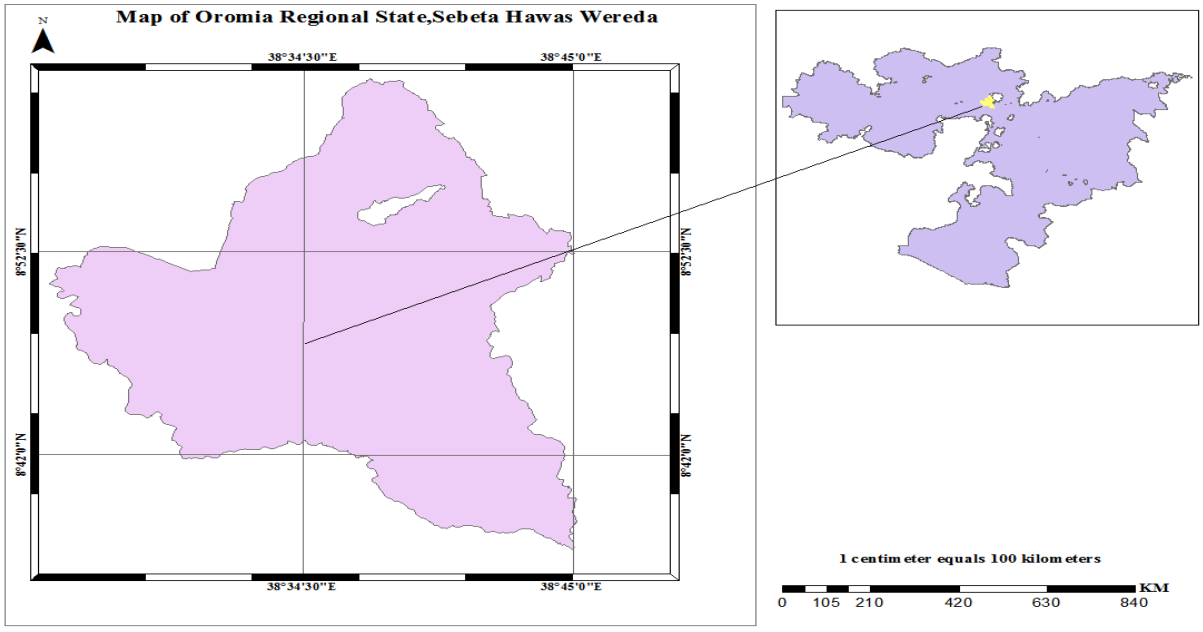

Figure 11: Sebeta district, North West Shoa zone, Oromia regional state, 2018.

2.4.2. Study design

2.4.3. The study design:

A retrospective study was conducted from previous data of malaria confirmed cases of Sebeta district.

2.4.4. Sampling technique

Purposively sampling from previous malaria weekly surveillance data pf PHEM from 2012-2017.

2.4.5 Data entry and analysis

Data collection, entry and analysis was carried out in Epi info 7.1 and excel software application and presented in tables and figures.

2.5. Result

During the study period (2012 -2017), the incidence of Malaria in Sebeta district health center was .The most frequent cases were registered in 2016, with the confirmed case of 8276(5.67%) followed by 2017 which is 7186(4.67%). However, the least positive cases were recorded in 2013 with the positive cases of 2989 (2.39%) as indicated in (Graph 12)

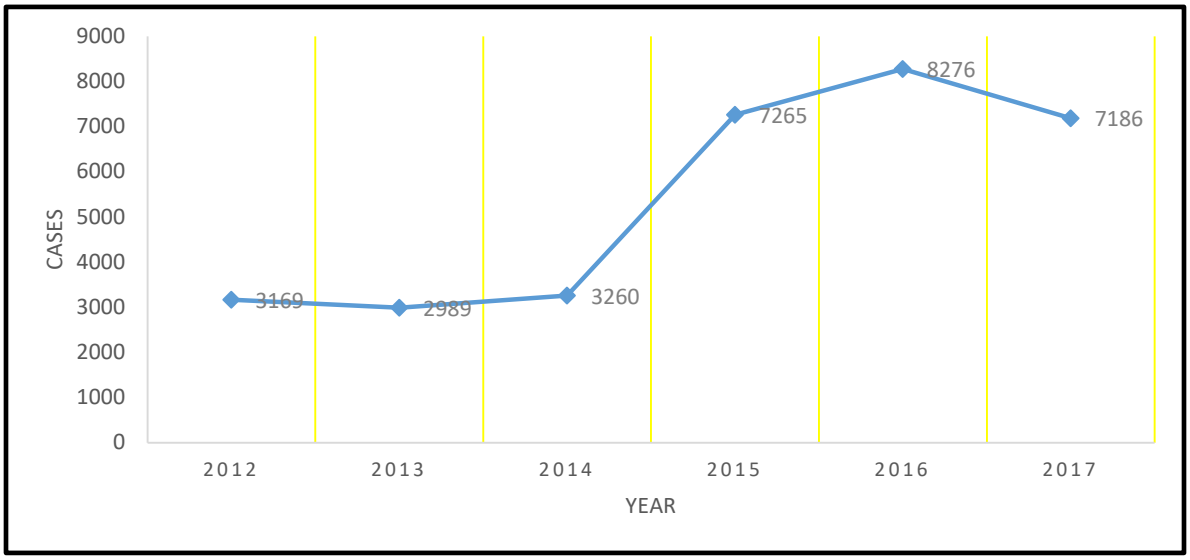

Figure 12: Malaria Trend of Examined Patients in Sebeta District from 2012-2017.

Regarding to the malaria species *P. vivax* was the more frequent than *P. falciparum*, which is about 60% of it. In both species the most frequent case was in 2016 followed by 2017 while the lowest frequently identified and confirmed positive cases in the year 2012. As in Figure 13.

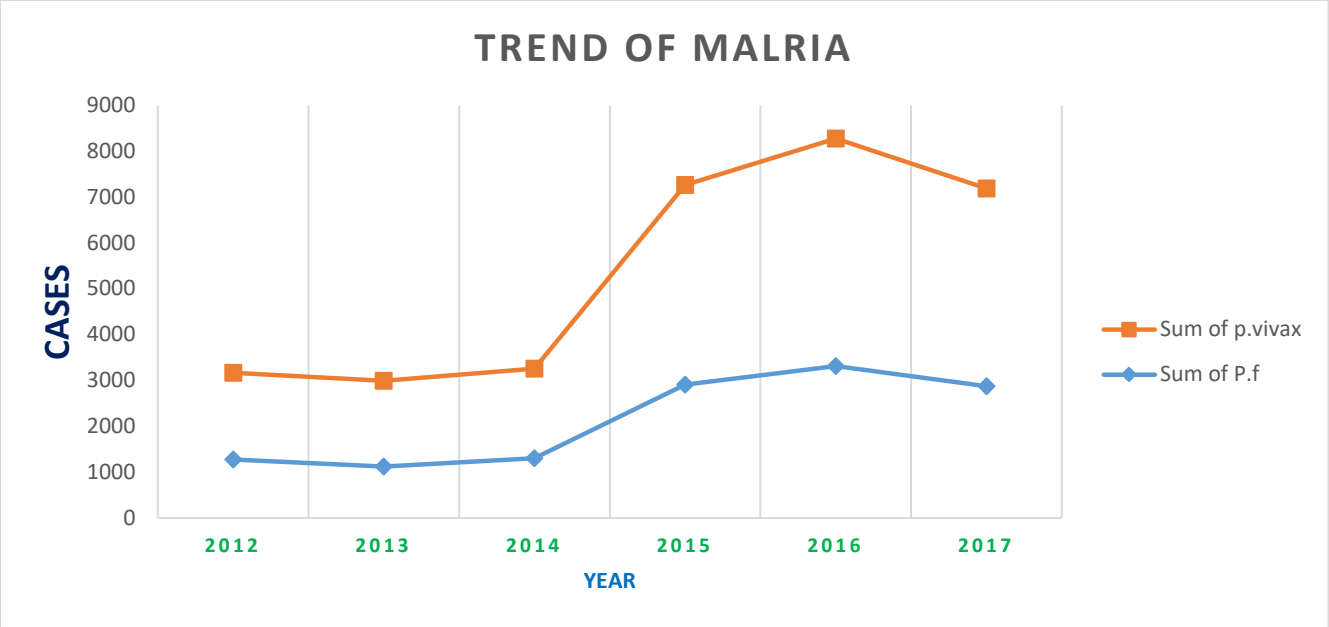

Figure 13: Trend of Malaria Species in Sebeta District, 2012-2017.

In comparison with the trend of the malaria confirmed cases regards to the species *p. vivax* (66.7%) is the most frequently occurred than *P. falciparum* (30.3%) relative to the total examined patients in over all the years of the trend from 2012-2017 as indicated in figure 14.

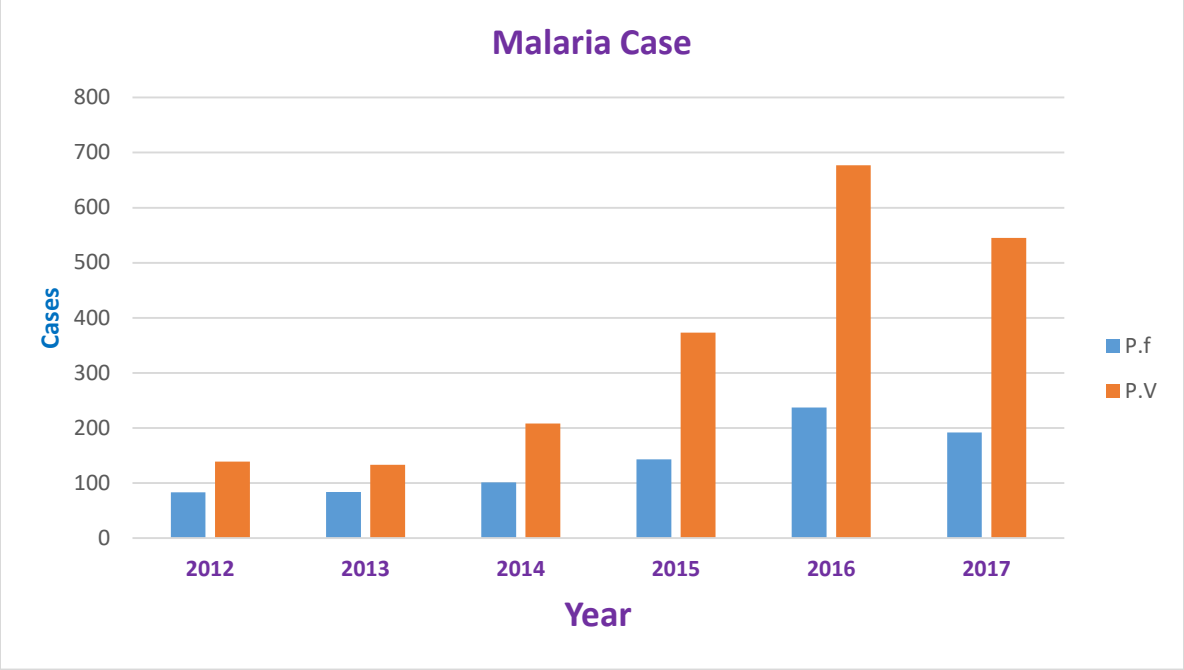

Figure 14: Malaria Confirmed Positive Cases In Species from the Year 2012-2018 of Sebeta Health Center.

In relation to the prevalence of Gender wise the most prevalently affected gender is female (67%) as shown in figure 15.

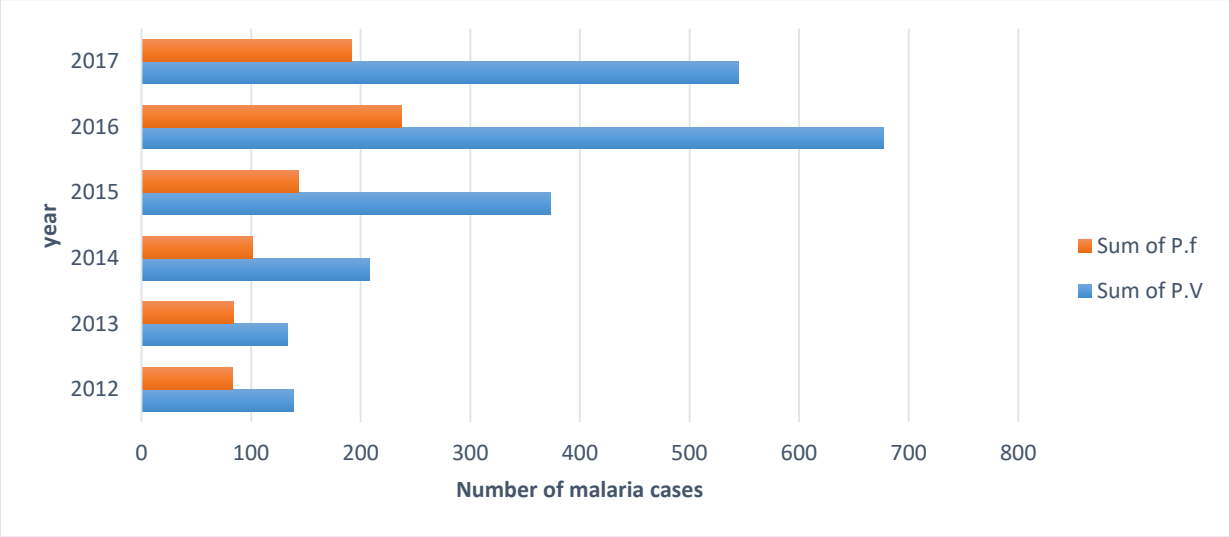

Figure 15: Incidence of Malaria Cases in Sex Wise of Year 2012-2017 from Sebeta Health Center

The Incidence of the malaria confirmed cases in relation to number of examined suspected cases the most prevalent year of the case is registered in year 2016 which is 5.67 % , year 2015 which is 5.24 % while the least of the prevalence registered in year 2013 as 2.39 % as indicated in Table 10.

Table 10: Distribution of Malaria Attack rate in percentage of Sebeta district from 2012 to 2017.

Year	Population	Case (N)	AR in Percent	Death	CFR
2012	118,828	3169	2.67	0	0
2013	125089	2989	2.39	0	0
2014	131664	3260	2.48	0	0
2015	138,595	7265	5.24	0	0
2016	145,889	8276	5.67	0	0
2017	153,567	7186	4.68	0	0

Regarding seasonal trend of the malaria distribution in 2016 of the highest trend among the years of 2012-2017, in months starts to incline from September to October the pick and decline from

November the list case detected from June at the beginning of the rainy season as indicated in fig 16.

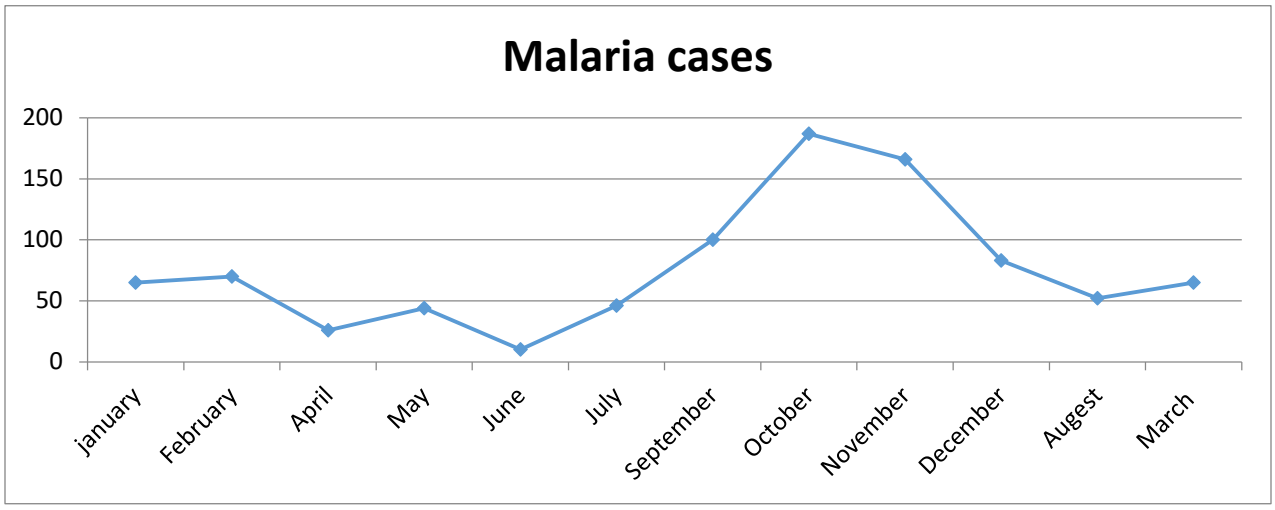

Figure 16: Trend of seasonal pattern in Sebeta district, 2018

The trend of Malaria in Sebeta district from 2012-2017 indicated that, there was similar pattern for three consecutive years 2012-2014. Started to rise from 2014 which showed doubling the situation in 2016 follow similar situation with slight difference in 2017. This might be due the improvement of the surveillance report of each case than the previous years.

The overall Attack rate of the malaria cases in relation to the population under risk of the health center catchment area shows 2.39-5.67 % and the most prevalent year in all species indicated in year 2016 and females were more infected than males. The plasmodium species that detected in the areas were p. vivax and p. falciparum with 66.7% and 33.3% respectively. =-087q u3; k

Unlike to our study, which is 60% of the cases was *P.vivax*, Malaria is a sever disease in Ethiopia, 75% of the land are malarias' areas and more than 54 million people are vulnerable. *Plasmodium falciparum* and *P. vivax* are commonly known species in Ethiopia are to cause malaria accounting for 60% and 40%, respectively. [15]. Since Sebeta district is high land area there might be relapsing case and re-examined of *P.vivax* species, while the *P. falciparum* species were immigrant people who stay temporary in Sebeta town being infected from Malaria endemic area like Gambella region while visited the healthy facility of the town.

Approximately 52 million people (68%) live in malaria risk areas in Ethiopia, primarily at altitudes below 2,000 meters. Malaria is mainly seasonal with unstable transmission in the highland fringe areas and of relatively longer transmission duration in lowland areas, river basins and valleys. Historically, there have been an estimated 10 million clinical malaria cases annually. Since 2006, however, cases have reduced substantially. On average, 60%-70% of malaria cases have been due to *P. falciparum*, with the remainder caused by *P. vivax*. *Anopheles arabiensis* is the main malaria vector; *An. pharoensis*, *An. funestus* and *An. nili* play a role as secondary vectors. [16]

Approximately 4-5 million cases of malaria are reported annually in Ethiopia and the disease is prevalent in 75 per cent of the country, putting over 50 million people at risk. Malaria accounts for seven per cent of outpatient visits and represents the largest single cause of morbidity. It is estimated that only 20 per cent of children under five years of age that contract malaria are treated at existing health facilities. Below are feature stories related to malaria [17]

The burden of malaria has been increasing due to a combination of large population movements, increasing largescale epidemics, mixed infections of *Plasmodium vivax* and *P. falciparum*, increasing parasite resistance to malaria drugs, vector resistance to insecticides, low coverage of malaria prevention services, and general poverty. Outpatient consultations, inpatient admissions and all in-patient deaths have risen by 21-23% over the last five years. Overall, malaria accounts for approximately 17% of outpatient consultations, 15% of admissions and 29% of in-patient deaths. However, as 36% of the population is out of reach of the health service coverage, these figures may under represent the true situation.[5]

Due to the unstable and seasonal pattern of malaria transmission, the protective immunity of the population is generally low and all age groups are at risk of infection and disease. Some small-scale studies have documented on malaria parasite prevalence between 10.4–13.5% in Gambella; 7.6–14.1% in Tigray; 4.6% in Amhara, 0.9% in Oromia and 5.4% in Southern Nations, Nationalities and People's Region in all age groups [18]

As the nation, the regions, and sub-regions seek to reduce malaria transmission to zero, the very high coverage (seeking universal coverage) of prevention interventions (LLINs, IRS) will serve to limit transmission in communities and will mean that there is very little potential for "epidemics" or "outbreaks"; that is, there is very low likelihood that a single introduced infection would expand to many infections/cases. Thus, the national emphasis on further reducing transmission will rely

on the existing prevention coverage and additionally focus on “surveillance” – the process of finding of individual human cases, treating and performing case investigation to identify the source and possible spread, with the aim of preventing any further malaria transmission. Therefore, this NSP aims to achieve a high quality, broadly based malaria infection detection, investigation and response ‘Surveillance System’ to further reduce malaria transmission and improve the detection and timely response to malaria epidemics. [19]

Conclusion and recommendation

The study result showed us there is steady statues trend of the malaria incidence for consecutive years 2012-2014 gradually from year 2014 stared to increase and pick in 2016 in next year similar trend was detected with slight lower.

Based on the result of the study I strongly recommend to Sebeta health center to pay attention the trend of malaria from year to year to take an action by improving surveillance system.

Since the incidence rate as shown from the result above 5% there should be through examination and integrated control measure has to be taken in all heath facilities and as a district.

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CHAPTER 3. HEALTH PROFILE ASSESSMENT

3.1. Health Profile Assessment Report in Sebeta district, South West Shoa Zone, Oromia Regional State, March 10-15, 2018.

Summary

Background

Sebeta district is located in south West part of Addis Ababa has a latitude and longitude of 8^0 54' 40" N 38^0 37' 17" E and an elevation of 2,356 meters (7,730 feet) above sea level. The six largest ethnic groups reported in Sebeta were the Oromo (67.37%), the Amhara (17.91%) the Sebat Bet Gurage (4.54%), the Sodo Gurage (4.43%) the Werji (2.5%) and the Gamo (1.02%) all other ethnic groups made up 2.23% of the population. According to the district Agricultural and Rural Development Office, 87.2% of the land is devoted to agriculture, 4.2% is pasture, 2.9% is forest, 1.86% is reserved for industrial establishment, 1.68% is covered by lakes and other bodies of water, and built-up land covers 1.28%.

Objective

The main objective this study is to determine the current status of the district, to know the main health related indicators and to describe the updated situation of public health profile.

Methods:

The study conducted cross-sectional study with pre-designed structured questionnaires and data was collected among the selected institution, raw data collection and analysis was made in to MS excel and Epi info 7.1 software application.

Results:

A cross-sectional study conducted in Sebeta district, out of the total 153,612 population majority of them under 25 years, 51% and 49% of female and male consecutively. There are about 143 health workers in 4 health center, The highest proportion of health worker to population ratio is urban health extension 1:4800, there is no any General practitioner in the district, MPH 1:38404, HO 1:8084. One health center serve for 38403 people. In contraceptive acceptance rate the most widely used method is injectable (78%), oral (15%), implant (3.4%) and the least IUCD (0.56%), 20-24(43.49%) year of age, 24-49(33.02%) the least under 15(0.81%). Water supply coverage comprises only 37%, electricity 60%, Immunization coverage above 100%. Proportion of School

coverage in KG and primary school class to number of students 1:46 where as in high school1:42. The sanitation coverage is below 15%. Woman of age 15-49 years are 22.13%, pregnancy (3.4%), NPW (18.63%), percentage of lactating woman (18.63%). Out of the total delivery woman who attended delivery in skill birth (97%) an average family size is about 4.8.

Conclusion

In Sebeta district there is low coverage of water supply, sanitation coverage, poor waste disposal management, and low proportion of health worker in comparative to total population in number and both in quality. Those indicated low coverage are the vital indicator for the improvement of the communities' health.

3.1.1. INTRODUCTION

Despite major strides to improve the health of the population in the last one and half decades, Ethiopia's population still face a high rate of morbidity and mortality and the health status remains relatively poor. Vital health indicators from the DHS 2005 show a life expectancy of 54 years (53.4 years for male and 55.4 for female), and an IMR of 77/1000. Under-five mortality rate has been reduced to 101/1000 in 2010v. Although the rates have declined in the past 15 years, these are still very high levels.[1]

There is a need by the health sector of developing countries to use the limited resource effectively in order to provide an efficient and equitable health service to the communities. This then requires sound management that is based on information, which is crucial at each level of the health service management.[2]

Correct and up-to-date information is critical, not only for the provision of high-quality clinical care, but also for continuing health care, maintaining health care at an optimal level, clinical and health service research, and planning and management of health systems.[3]

The main objective of the profile assessment is to know the current status of the district with health and related facilities that directly or indirectly contribute for the daily life of the community.

3.1.2. OBJECTIVE

General Objective:

The main objective of the public health assessment is to know the current status of health and health related facilities in Sebeta district.

Specific Objectives:

- To describe the current status of health profile
- To describe up to date information regards to health and health related facilities in Sebeta district

3.1.3. LITERATURE REVIEW

Ethiopia is Africa's oldest independent country. It is the tenth largest country in Africa, covering 1,104,300 square kilometers (with 1 million sq. km land area and 104,300 sq km water) and is the major constituent of the landmass known as the Horn of Africa. It is bordered on the north-northeast by Eritrea, on the east by Djibouti and Somalia, on the south by Kenya, and on the west-southwest by Sudan. Its geographical coordinates are between 8 00 N and 38 00 E.

The population age pyramid has remained predominately young: 44% are under 15 years, over half (52%) are between 15 to 65 years, and only 3% of all persons are over the age of 65 years. The sex ratio between male and female is almost equal, and women in the reproductive ages constitute 24% of the population. While the average lifetime fertility has declined in the past 15 years from a 1990 level of 6.4 births per woman down to 5.4 births [4] rural women have an average of three more births per woman compared to women in urban areas. Overall, even with the fertility decline, the population is still growing at an annual rate of 2.6%. The lowest rate of population growth is in the Amhara region, which, at 1.7%, is lower than population replacement.

The Federal Democratic Republic of Ethiopia has nine Regional States: Tigray, Afar, Amhara, Benishangul-Gumuz, Gambella, Harari, Oromia, Somali, and Southern Nation Nationalities and Peoples Region (SNNPR); and the City Administration Councils of two cities: Diredawa and Addis Ababa. The regional states and city administrations are subdivided into 817 administrative Woreda's (districts). A Woreda/District is the basic decentralized administrative unit and has an administrative council composed of elected members. The 817 Woreda's are further divided into about 16,253 Kebeles, the smallest administrative unit. There are also two zones and seven Woreda's designated as "special". These are medium sized towns or traditional sites of various population groups.

The health and wellbeing of any population requires a holistic approach that includes the involvement of many agencies and gives ownership to the communities involved. The traditional notion of top-down delivery of health care is no longer acceptable to central governments, who are

seeking greater value for money. The increasing return to the principles of public health signifies that a purely medical approach to health cannot by itself resolve the many health problems in increasingly complex cities. Government health strategy documents increasingly recognize the importance of the views of the people receiving services in needs-based service delivery and espouse the involvement of individuals and communities as a key objective in the future delivery of health services[5]

Ethiopia has a poor health status in relation to other low-income countries, even within Sub-Saharan Africa (Health, E.M.o. *Overview of the Sector*. 2013). The most serious global health problems are in poorer countries, and good health information is vital in tackling these problems. Effective HIS offer government and health department officials a clearer understanding of the effects of their policies on the health of their people [6]

Ethiopia's economy depends heavily on the agricultural sector; agriculture accounts for 83.4% of the labor force, about 43.2% of the Gross Domestic Product (GDP) and 80% of exports7]. Regular droughts combined with poor cultivation practices, make Ethiopia's economy vulnerable to climatic changes. Despite such obvious challenges, Ethiopia has shown an impressive economic growth over the last seven years.

The Poverty Head Count Index has declined from the 1996 level of 45.5% to 32.7% in 2007/08 [7]. The reduction in poverty has been more pronounced in rural than urban areas. Even with this growth, the annual per capita earnings of 235 USD remain below the Sub-Saharan average. The overall economic dependency ratio for the country is estimated at 93 dependents per 100 persons in the working age group of 15-64 years.

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Considering the HRH staffing numbers and composition required to achieve universal Primary Health Care (PHC) coverage by the end of HSDP III, the FMoH focused on scaling up training of community and Mid-Level Health Professionals (MLHPs). To meet the needs for the community level HEP, a total of 31,831 HEWs were trained and deployed. For MLHPs, an Accelerated Health Officer Training Program (AHOTP) was launched in 2005 to address the clinical service and public health sector management needs at district level.

So far, more than 5,000 health officer trainees (generic and upgrade) have been enrolled with 3,573 Health officers graduated and deployed. To address the HRH need for Comprehensive Emergency Obstetric Care (CEmONC) and other emergency surgical services at PHC level, a masters' level curriculum on Emergency Surgery was developed and training started in five universities. To address critical shortages and the mal distribution of doctors, a new medical school was opened in St. Paul's Hospital, the Millennium Medical School. This school has an innovative curriculum that integrates clinical skills and social accountability.

3.1.4. METHODOLOGY

3.1.4.1. Study area

The Study area was conducted in Sebeta district, South west part of Ethiopia Located in Oromia special zone surrounding Finfinnee, of the Oromia region about 27 km from Addis Ababa, The district has an area 87,532 hectare, an altitudinal range of 1800-3385 masl, The district is divided in to highland (12%) and mid highland(88%). Sebeta town located in 100 km from zonal office.

3.1.4.2. Data collection: Pre-designed closed questioner was distributed to all stakeholders of concerned institutes and organizations and data entry and analysis was done in excel sheet and epi info 7.1 Software application.

3.1.4.3. Study design: the study was conducted in cross-sectional descriptive study the result illustrated in tables graph

3.1.5. RESULT

Sebeta district has four governmental health center namely: Sebeta, Daleti, Alemgena and wolette HCS the location of Sebeta is west part of Finfinnee which is 27 km from the center.

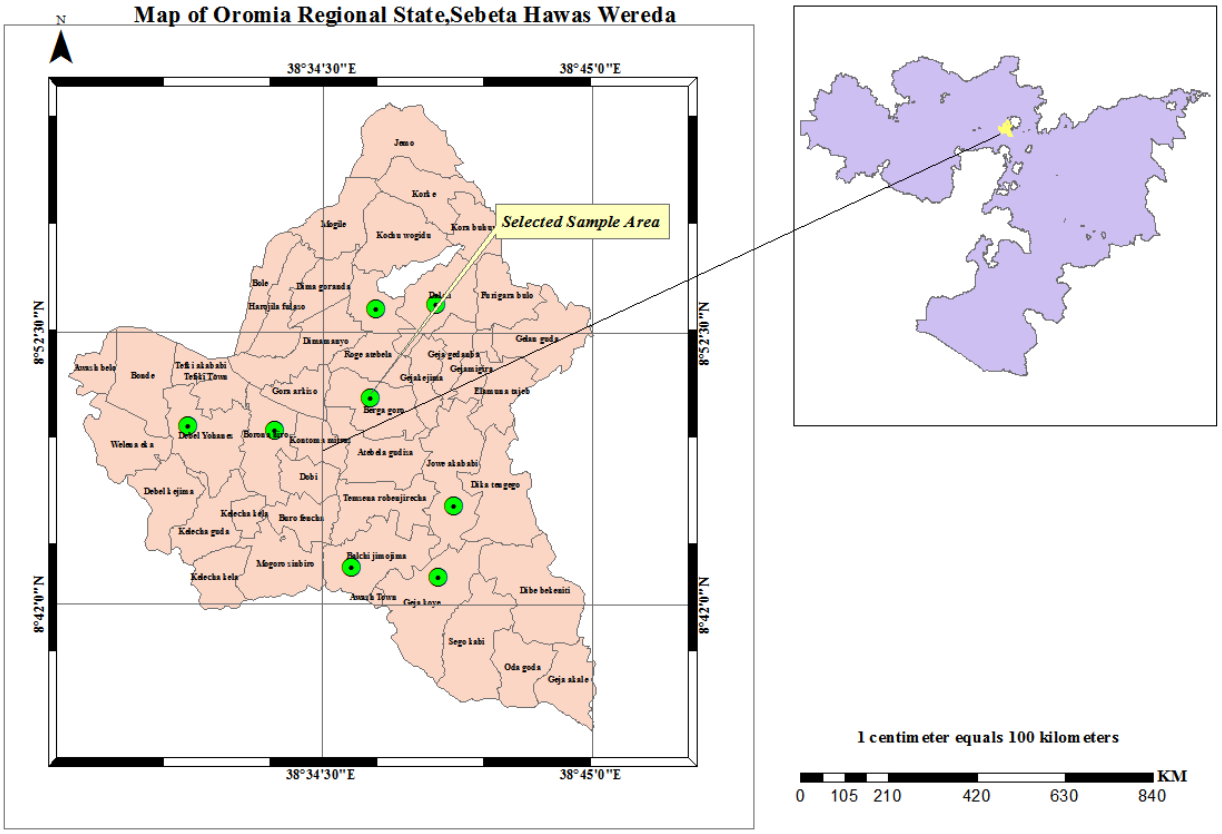

Figure 17: Sebeta district and location of health centers, 2018

Demography

Demography is one of the most important variable to determine the vital statistics, mortality rate GDP, and major infrastructural coverage. Population dynamicity of the Sebeta district in age category and male to female proportion ratio indicated in 2018 majority of the population is under

age of 5, 30183 (19.65%) followed with an age of 16-25,24235 (15.78%) the least number of the population is above 65, 2300 (1.5%). Table 11.

Table 11: Distribution of Population in Sebeta District with Age Category, 2018.

Age Category	**Male**	**(%)**	**Female**	**(%)**	**Total**	**(%)**
<5 year	17808	11.59	12375	8.06	30183	19.65
6-15 year	6684	4.35	13455	8.76	20139	13.11
16-25 year	10046	6.54	14189	9.24	24235	15.78
26-35 year	7174	4.67	14316	9.32	21490	13.99
36-45 year	12473	8.12	8315	5.41	20788	13.53
46-55 year	14689	9.56	8800	5.73	23489	15.29
56-65 year	5212	3.39	5776	3.76	10988	7.15
>65 year	1341	0.87	959	0.62	2300	1.50
Total	**75427**	**49.10**	**78185**	**50.90**	**153612**	**100**

In regarding Gender distribution of the total population majority of the people in all age groups are Female (50.9%). Fig 18.

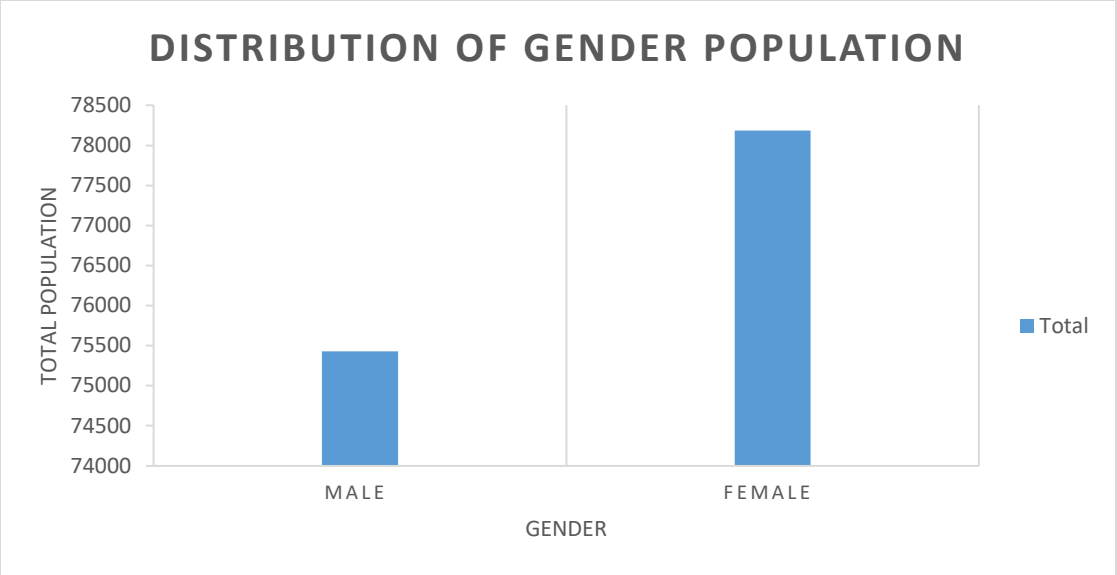

Figure 18: Population Distribution of Sebeta District In, 2018

In Age group distribution of the district majority of the people who is living in the district is an age under five 30177(19.67%) out of this (11.5%) is male and (8.6%) is female followed by 16-25 years 24235 (15.77%) in regarding to gender Male 6.54 % female (9.24%) from the least age distribution above 65 (1.5%) sex distribution (0.87%) and (0.62%) male and female respectively. Figure 19.

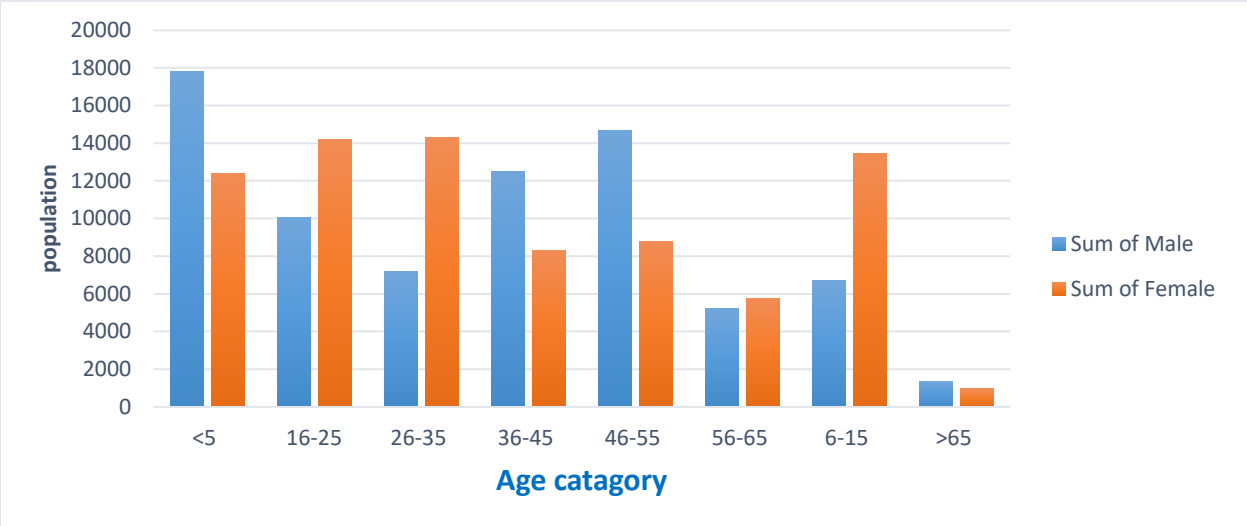

Figure 19: Population Distribution with Age and Sex of Sebeta District in 2018.

Education

The profile of the educational background and facilities there are 100 KG and primary school, 12 secondary school and 2 collage of formal education both in government and private Schools exist in the district, with regards to teachers to student ratio 1:46 in KG and Primary School and 1:42 in secondary high school as indicated in Figure 21.

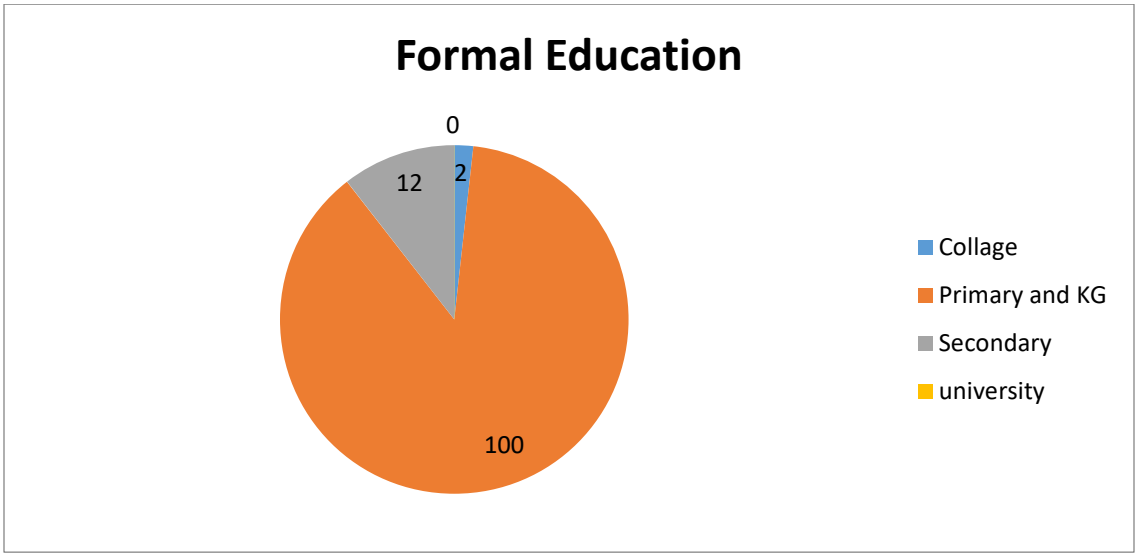

Figure 20: Distribution of Formal Education in Sebeta District In, 2018

Retrospective study from 20014-2017, the trend of School drop out of the students in KG, Primary and High school for the past five years the largest number was in 2017 followed by 2015 and the least number of student who dropout during the study was in 2014 as indicated in figure 21.

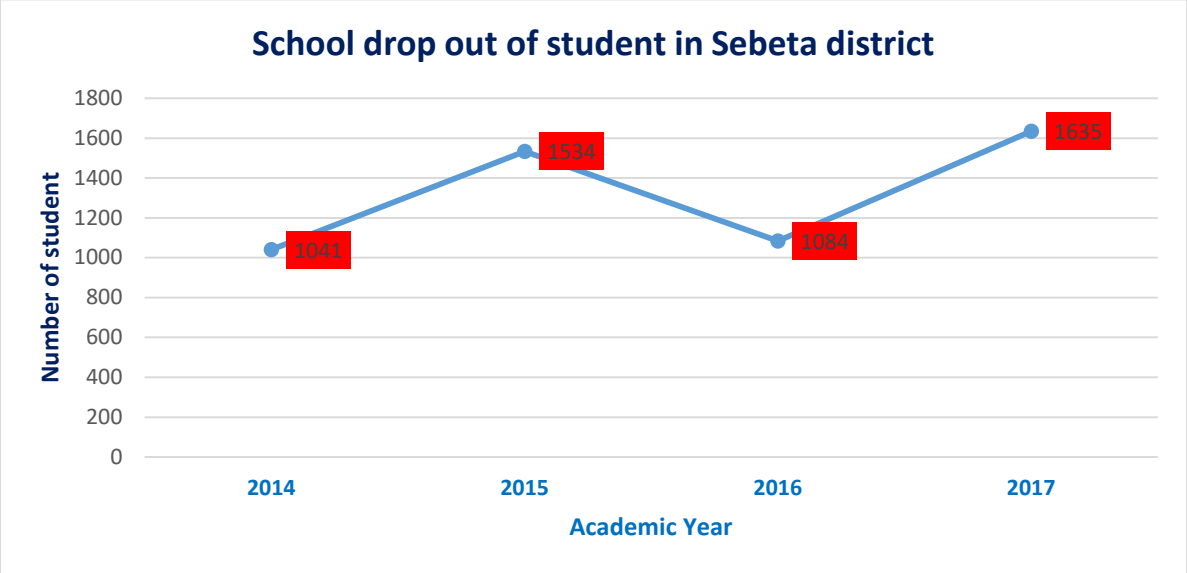

Figure 21: Trend of School Dropout Student in All Level of Formal Education, 2014-2018 in Sebeta District.

The four years school dropout from KG and Primary School due to different reasons the highest number of students who dropout school is in year 2017 (1437) out of those female are (51%) and Male (49%) followed by 2015 (1357) student of which (51.4%) Male and (48.4%) were Female students.

As the result indicate that from Secondary School the highest number of student dropout was similar to KG and primary School in year 2017 (198) of which(53%) Female and (47%) were male students, followed by 2015 which is 177 students out of which (60%) male and (40%) Female students as indicated in Figure 22.

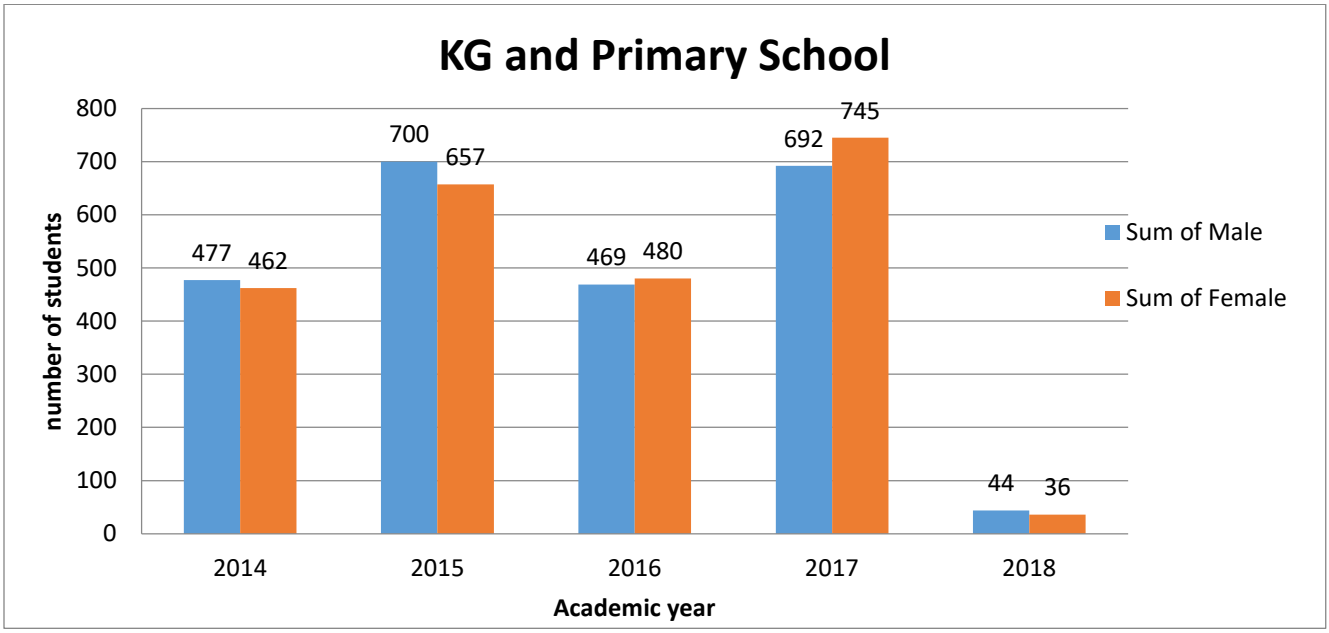

Figure 22: School Dropout of Student from Kg and Primary School from 2014-2018 In Sebeta District.

The school dropped out students from high-school level the highest number who drop school is in year 2016 male students followed with 2017 female student while the least in 2018 in both sexes. As indicated in figure 23.

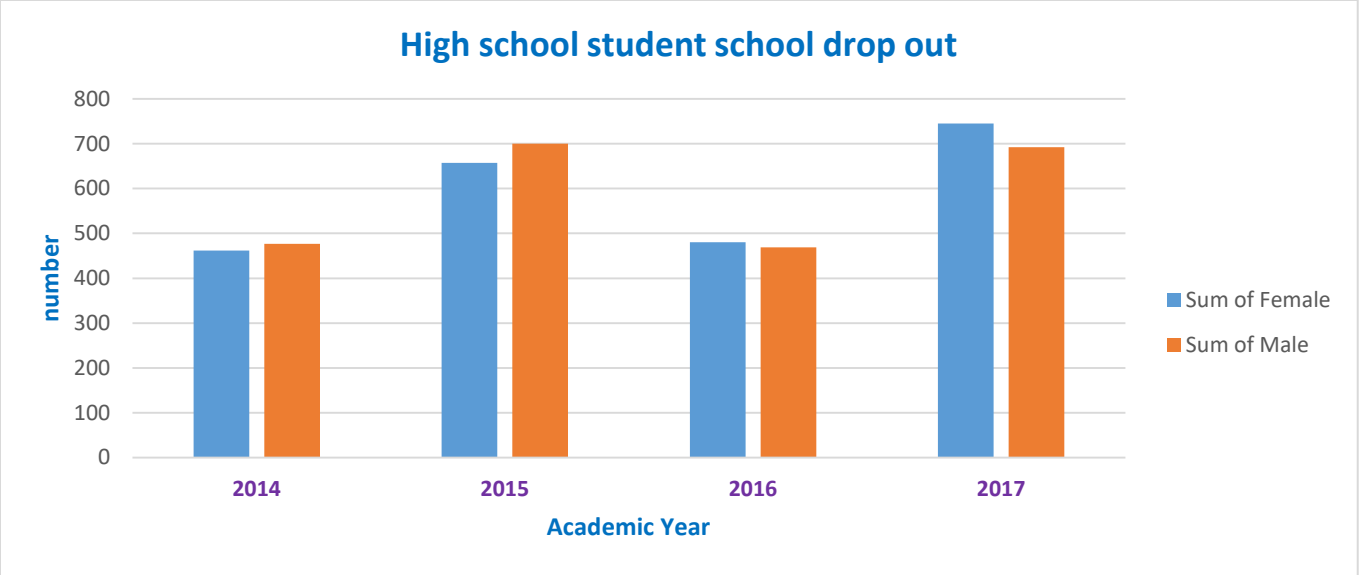

Figure 23: Distribution of School Drop Out Of Students from High School in Sebeta District 2014-2017

Health Facility

Health facility is one of the millennium goal for the reduction of Maternal and early delivered infants as a result of this there are several health facilities were constructed and some of them are under construction and there are different opportunities to upgrade the health workers in different level of qualification and number to achieve this goal. With regards to man power with in Sebeta health center there are 193 workers of different level as indicated in Table 12.

Table 12: Distribution of Health professionals in all Health Centers of Sebeta district in 2018

S/N	Qualification	Male(n) %	Female(n) %	Total
1	Second degree			
1.1	MPH	3(75%)	1(25%)	4(100%)
2	First degree			
2.1	General practitioner	0	0	0
2.2	Health officer	5(26%)	14(74%)	19(100%)
2.3	BSC Nurse	2(9%)	12(81%)	22(100%)
2.4	BSC Midwifery	2(100%)	0	2(100%)
2.5	Laboratory technologist	2(50%)	2(50%)	4(100%)
2.6	Environmental Health	4(80%)	1(20%)	5(100%)
2.7	Pharmacist	4(50%)	4(50%)	8(100%)
2.8	Health Education & promotion	2(100%)	0	2(100%
3	Diploma			
3.1	Clinical Nurse	5(28%)	18(72%)	23(100%)
3.3	Midwifery nurse	1(10%)	9(90%)	10(100%)
3.4	Druggist	2(67%)	1(33%)	3(100%)

The healthy coverage of Sebeta health centers is 100%, from existing facilities, there are about 131 health facilities of different level and function both in clinic and pharmacy in Government and non-government private, NGOs and owned by different Factories, out of those facilities there are only 4 health center and four pharmacies of governmental health facility as indicated in Figure 24.

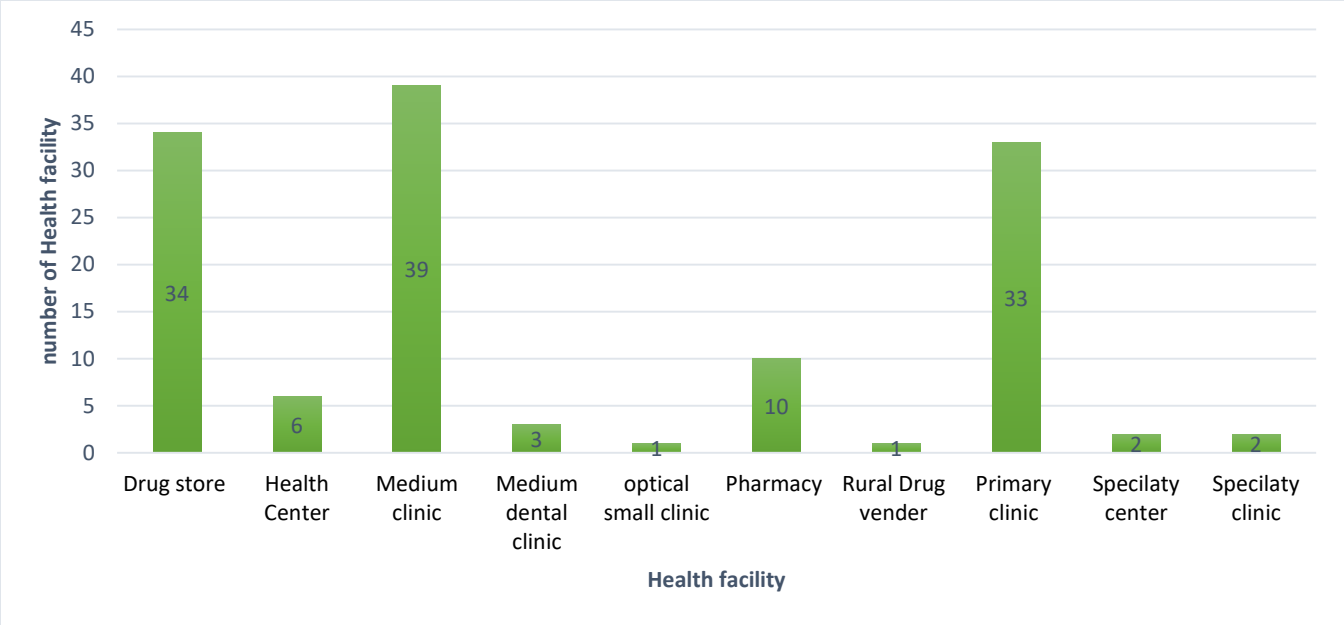

Figure 24*:* Distribution of Health Facilities of Sebeta District In, 2018

For the Health facilities, the health worker in different level of qualification indicated majority of the health worker is lower level of educational background who had Diploma level Urban Health Extension (25.2 %) followed by clinical nurse (20 %) and the least professional health workers is General Practitioner, Health education and Promotion and MPH (0.86%) each. From all health workers and average health worker to population 1:1172 in comparison of each profession to population ratio MPH 1: 76803, HO 1:25601, General practitioner 1:153606 and BSC Midwifery 1:153606 which is very significant number as indicated in Figure 25.

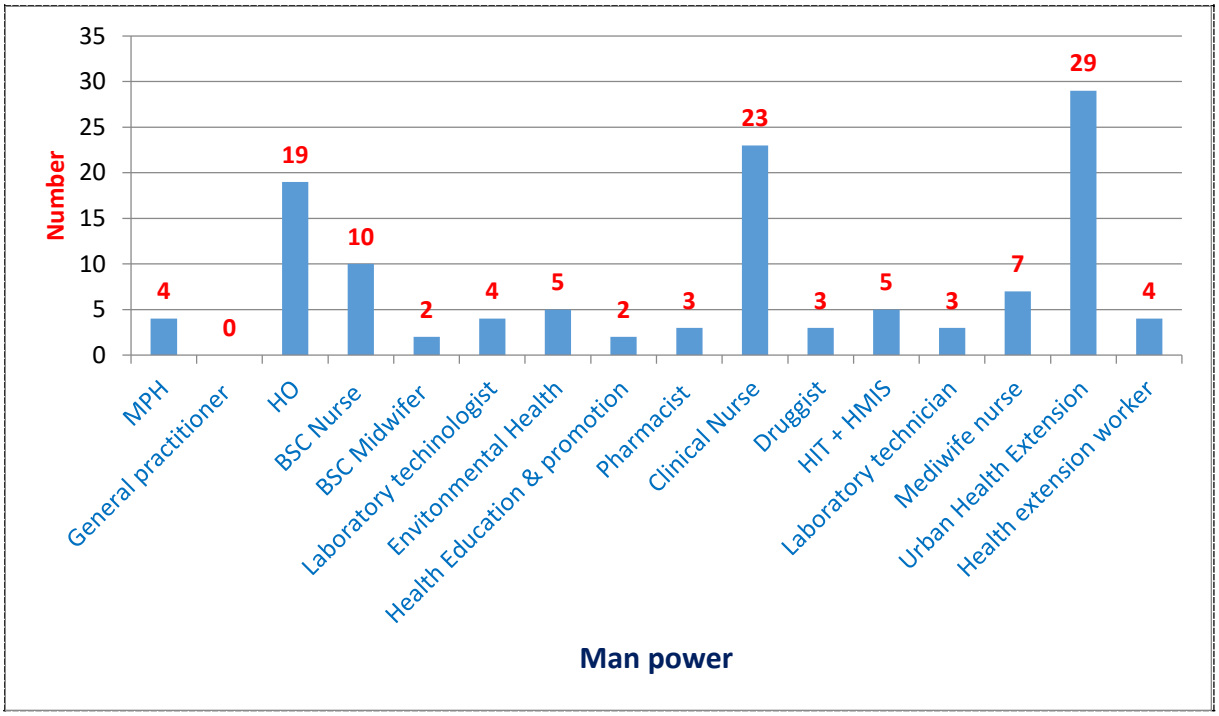

Figure 25: Distribution of Man Power in Sebeta Health Facilities In, 2018.

Immunization

Immunization in Sebeta district health centers indicated the highest number of immunization is TTI followed by Rota II while the least immunization is BCG as indicated figure 26.

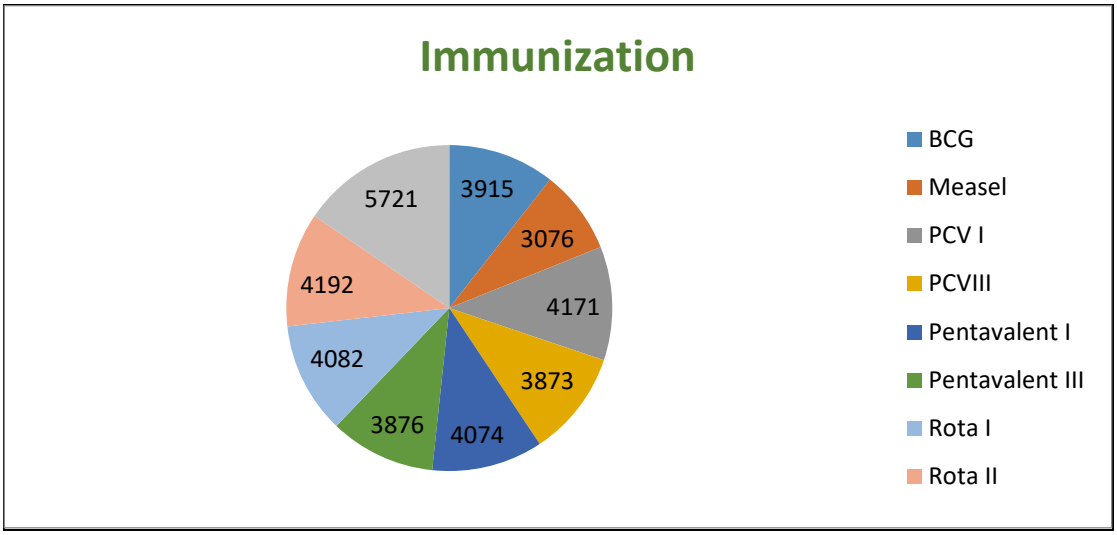

Figure 26: Immunization Coverage of Sebeta Health Center, 2018.

Vital Statistics

From vital statistics from total population woman of age 15-49 years are 22.13%, pregnancy (3.4%), NPW (18.63%), percentage of lactating woman (18.63%). Out of the total delivery woman who attended delivery in skill birth (97%) an average family size is about 4.8.

Pregnant woman antennal natal care (ANC) and post-natal care (PNC) trend health centers increasing gradually from year to year, as indicated in Table 12.

Table 13: Antenatal and Postnatal Care Of Sebeta Health Centers from 2015-2017

S/N	Ante and post Natal care	2015	2016	2017
1	ANC at least once	6720	7967	8697
2	ANC at least four times	2488	3706	3228
3	Early PNC attendant 0-48 hrs.(0-2days)	1774	2004	2712
4	Early PNC attendant 49-72 hrs.(2-3 days)	54	229	141
5	Early PNC attendant 73 hrs. to 6 days(4-6 days)	237	722	398
6	Number of life birth	2447	3279	3888
7	Number of still birth	7	13	8

Contraceptive acceptance rate in health centers, out of used methods are oral, injectable implants, IUCD and others, out of those methods the most accepted method is injectable in comparing four year retrospective data followed by oral and the least accepted rate is IUCD as indicated in Figure 27.

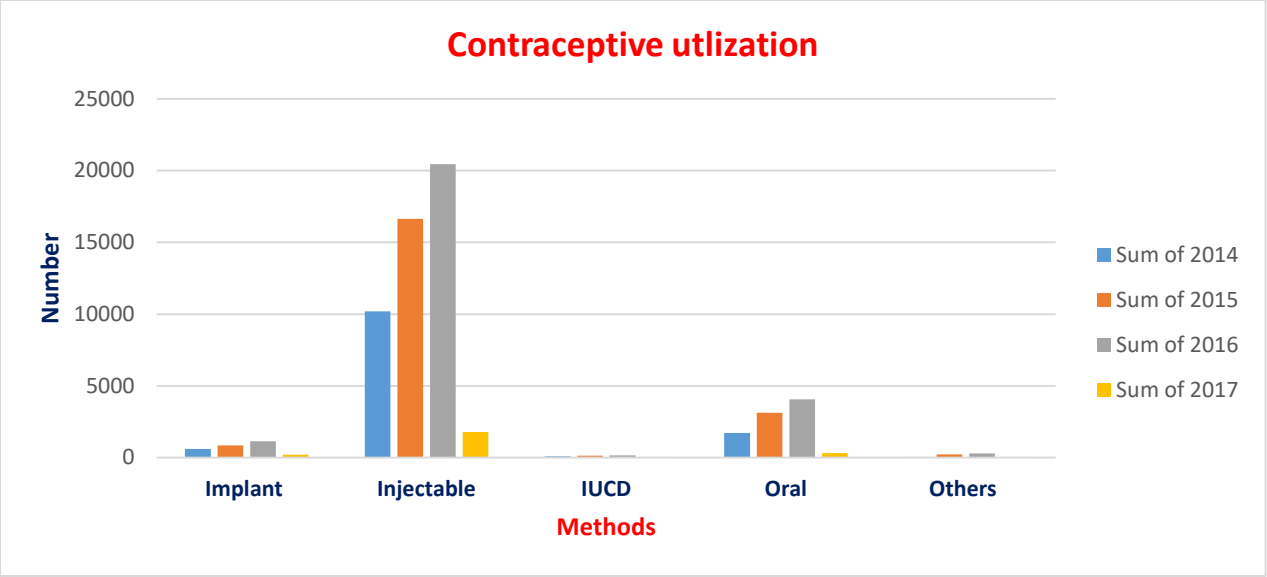

Figure 27: Contraceptive Acceptance Rate By Method in Sebeta Health Centers from 2014-2017

As indicated in Fig.29. Out of newly accepted contraceptive during 2015-2018 indicated that gradually increased from year to year, whereas in related to age category acceptance rate is an age of 20-24 is the highest followed by 24-49 years of age where as the least is under 15 age during the study period.

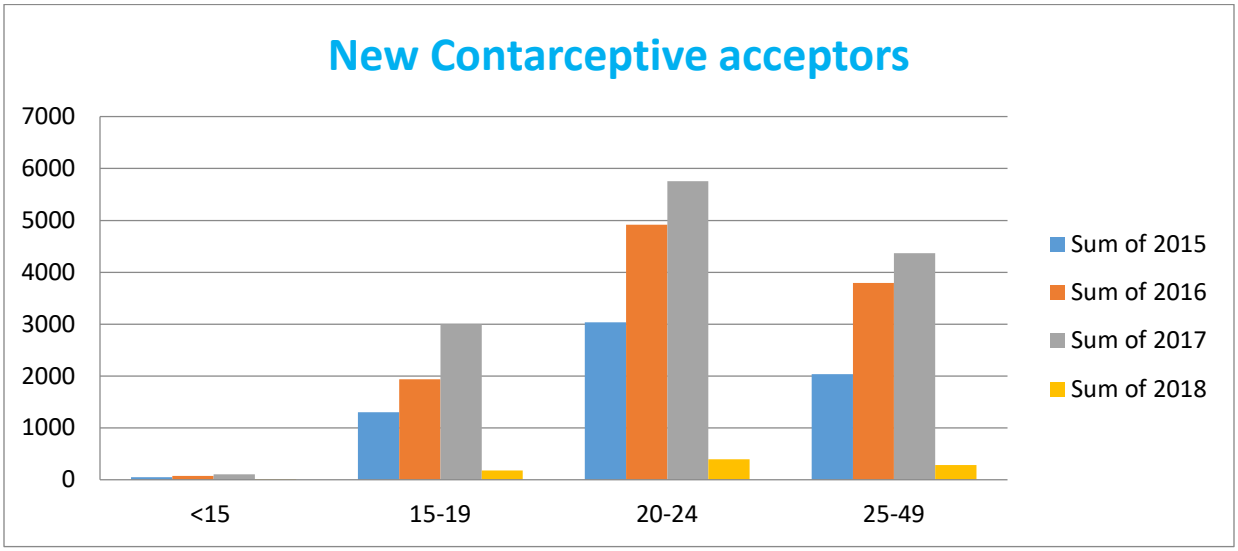

Figure 28: Age Distribution of Contraceptive Acceptance Rate in Sebeta Health Centers from 2015-2018.

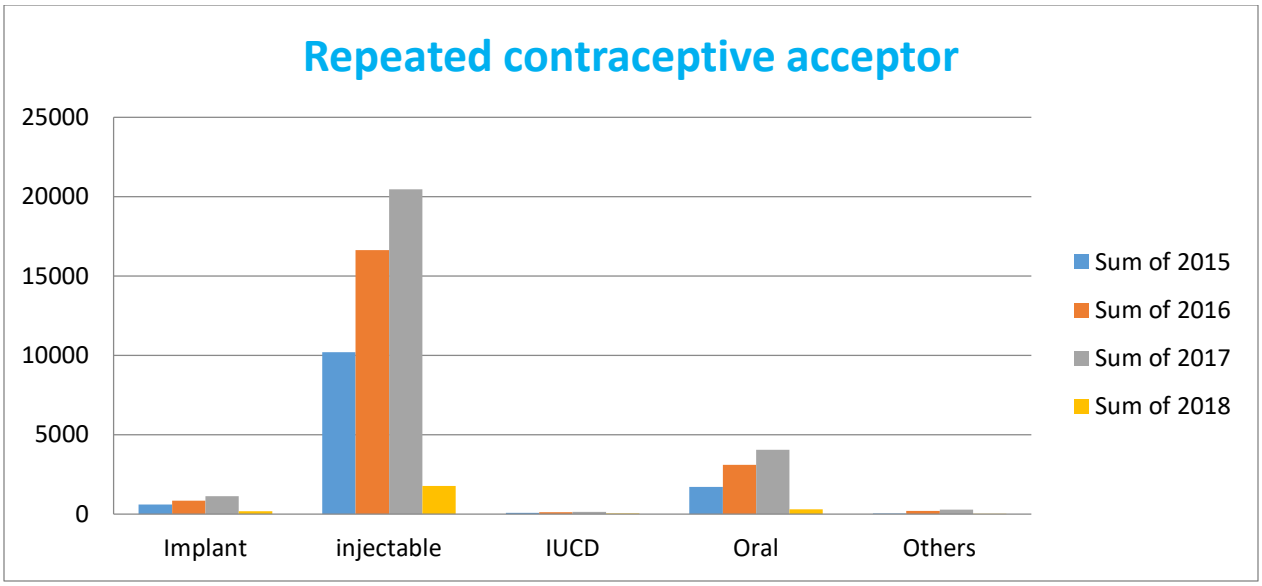

Figure 29: Distribution of Contraceptive Repeated Acceptor in Sebeta Health Center from 2015-2018.

Out of new delivered live birth screened the health facilities recorded data indicated that about 26% nutritional status is moderately mal nourished, (2.6%) is severely mal nourished.

Top ten Couse of Adult Female in Sebeta district, acute respiratory infection (ARI) is the leading Couse followed Diarrhea (non-bloody*)* and pneumonia is the least Couse as indicated in table 14.

Table 14: Top Ten Couse Morbidity of Female in Sebeta district, 2018.

S/N	Causes	Cases(N)	People affected in (%)
1	Acute upper respiratory infections	17055	24.3
2	Diarrhea (non-bloody)	8337	11.88
3	Acute Febrile Illness (AFI)	7818	11.44
4	Urinary Tract Infection(UTI)	7745	11.04
5	Typhoid Fever	7553	10.76
6	Dyspepsia	7060	10.06
7	Infections of the skin and subcutaneous tissue	4557	6.48
8	Disease of Musculoskeletal system and connective tissue	3701	5.27
9	Trauma (injury, fracture etc.)	3487	4.97
10	Pneumonia	2865	4.08

Acute Upper respiratory infection is the leading Couse among top ten morbidity16453 (28.98%) in Male, while acute bronchitis is the least Couse 2507(3.96%) as indicated in Table 15.

Table 15: Top ten Couse Morbidity of Male in Sebeta district, 2018

S/N	Couse	Cases(N)	People affected in %
1	Acute upper respiratory infections	16453	25.98
2	Diarrhea (non-bloody)	9031	14.26
3	Acute Febrile Illness (AFI)	7659	12.10
4	Typhoid Fever	6769	10.69
5	Trauma (injury, fracture etc.)	5662	8.94
6	Dyspepsia	5098	8.05
7	Urinary tract infection	3674	5.80
8	Disease of Musculoskeletal system and connective tissue	3302	5.21

9	Pneumonia	3163	5.00
10	Acute bronchitis	2507	3.96

Top Five Couse of morbidity for under five years

Totally there were 18, 887 causes identified in top five, in under five years of age, from the top five Acute upper respiratory infections is the leading cause 9189(41.28%) followed by Diarrhea (non-bloody) 4716(21.19%), Acute Febrile illness(AFI) is the least, 1154(5.18%) as indicated in table 16.

Table 16: Top five Couse of morbidity under five, Sebeta district, Oromia regional state, 2018.

S/N	Couse	Cases(N)	People affected in %
1	Acute upper respiratory infections	9189	41.28
2	Diarrhea (non-bloody)	4716	21.19
3	Pneumonia	1922	8.63
4	Infections of skin and subcutaneous tissue	1906	8.56
5	Acute Febrile illness(AFI)	1154	5.18

Sanitation:

Sanitation coverage, as urbanization growth increase from year to year the number of people who inhabitant followed similar pattern, while the sanitation coverage from waste disposal, latrine coverage from an individual house to public service giver is, only 15% of the total population had sanitation coverage whereas, latrine coverage exceed to 45%. With regards to waste disposal management there is no any disposal management.

Water Supply:

Pipe water supply coverage for the community within the district during 2013-2017 indicates that in 2013(70.1%) is the highest coverage of all the years followed by 2016 (68.6%) and the least coverage is in 2017 (39%) Figure 30.

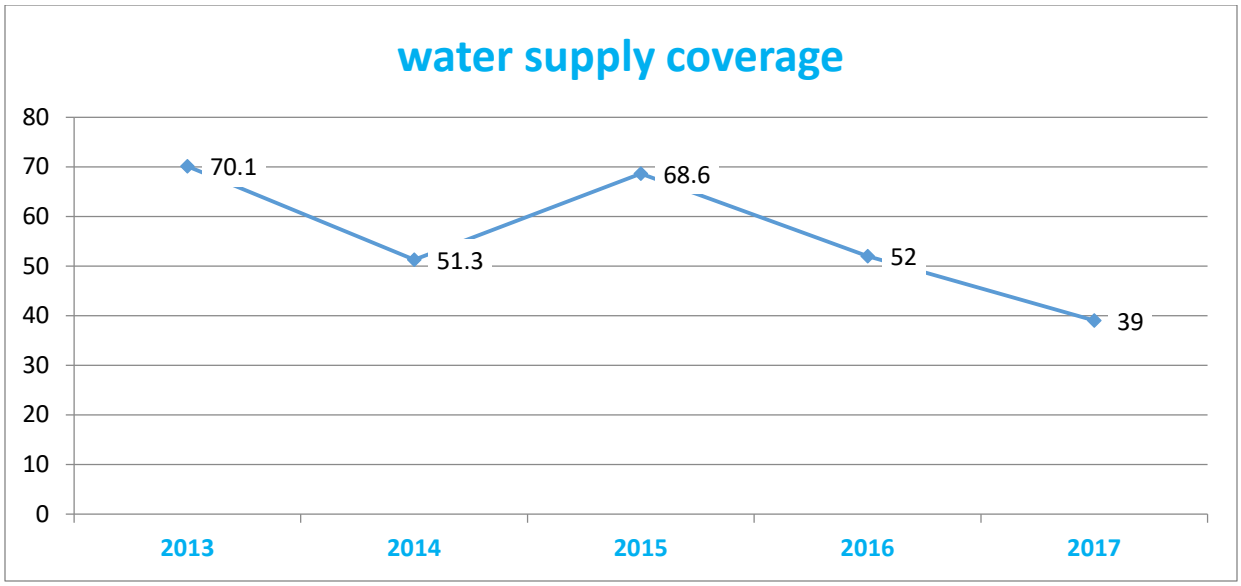

Figure 30: Pipe Line Water Supply Coverage of Sebeta District during 2013-2017

Electricity

As urbanization grows continuously as Electric power supply is one of the human basic needs, currently there is very huge number of inhabitants in need but as a coverage showed there about 16300 had individual electric power within the district, In comparison with the households an individual who had power source is 16,300(60 %).

Telecommunication:

Telecommunication coverage of the Community fixed line telephone within the district till 2018 is 3661 which shows about (13.53 %) from the total house hold population of the community living in the district. While the number of Cell phone is not included in the study because people who is living in the district had subscribed mobile phone service from different parts of the country.

Economy:

The economy of the district indicated that people who is living in the district had, government employee, private and other commercial activities like trade, Hotel, Factories. With regards to Government employee there are about 1409 out of which 774(59.93%) male and 635(45.06%) female employer with in Sebeta town. Range of monthly income is 688-10099 Eth Birr. Daily income of daily laborer 35-100 birr per day this income is not fixed based on the dealer of individual and type of task performed, monthly income of the factory worker is 650-1000 Eth. Birr.

Transportation

Public Transport facility of the district is 100% all weather road in all direction from the district to Addis Ababa and to district since the main road cross the town, the transport flow is passenger can access to public transport.

2.1.6. DISCUSSION

According to EDHS [8] study conducted the result indicated about 36 percent of currently married women are using a method of family planning: 35 percent are using a modern method, and 1 percent are using a traditional method. Among currently married women, the most popular methods are inject able (23 percent), implants (8 percent), IUD, and the pill (2 percent each). The contraceptive prevalence rate (CPR) among married women increases with age, peaking at age 25-29 (41 percent) before declining steadily to 19 percent among women age 45-49. Urban women are much more likely than their rural counterparts to use any method of contraception (52 percent versus 33 percent). By region, contraceptive prevalence rate ranges from 2 percent in Somali to 56 percent in Addis Ababa.

Contraceptive use increases with women's education and household wealth. For instance, 31 percent of women with no education are using a contraceptive method compared with 55 percent of women with more than a secondary education. Women with no living children (30 percent) and those with five or more children (28 percent) are the least likely to use any method of contraception.

Among sexually active unmarried women, 58 percent are currently using a contraceptive method: 55 percent are using a modern method and 3 percent are using a traditional method. The most commonly used methods among sexually active unmarried women are injectable (35 percent), implants (11 percent), the male condom, and emergency contraception (4 percent each).

According to EDHS study in 2011 showed, the rate of mortality associated with pregnancy and childbearing is 0.66 maternal deaths per 1,000 woman-years of exposure, down from 1.1. The estimated age-specific mortality rates display a plausible pattern, being generally higher during the peak childbearing ages than in the younger and older age groups. However, the age-specific pattern should be interpreted with caution because of the small number of events: only 118 maternal deaths among women of all ages. Maternal deaths represent 25 percent of all deaths among women age

15-49 during the 7-year period preceding the survey (118 maternal deaths divided by 473 female deaths).

As WHO study in 2010 indicated, as major strides to improve the health of the population in the last one and half decades, Ethiopia's population still face a high rate of morbidity and mortality and the health status remains relatively poor. Vital health indicators from the DHS 2005 show a life expectancy of 54 years (53.4 years for male and 55.4 for female), and an IMR of 77/1000. Under-five mortality rate has been reduced to 101/1000 in 2010v. Although the rates have declined in the past 15 years, these are still very high levels.

In terms of physical health facilities, 3,135 new Health Posts were constructed, far exceeding the insubstantial amount of 76 HPs in 1996/97. Health Centers were increased from the number in 1996/07 of 243 to 519 in 2003/04; hospitals were also increased from 87 in 1996/97 to 126 in 2003/04.[9]

Similar to my study result, the study result of EDHS 2011 indicated, nationally, the proportion of Ethiopian households with access to piped water has increased from 18 percent in 2000 to 24 percent in 2005 and 34 percent in 2011. In the last six years there has been a rapid increase in the percentage of households in Ethiopia that use some type of improved source of drinking water, from 35 percent in the 2005 EDHS to 54 percent in the 2011.

According to WHO and UNICEF study conducted in 2010, as ensuring adequate sanitation facilities is another Millennium Development Goal that Ethiopia shares with other countries. At the household level, adequate sanitation facilities include an improved toilet and disposal that separates waste from human contact. A household is classified as having an improved toilet if it is used only by members of one household (that is, it is not shared) and if the facility used by the household separates the waste from human contact [10].

Only about one household in every four (23 percent) has electricity, with a very large disparity between urban and rural households (85 percent versus 5 percent). In urban areas the proportion of households with electricity rose from 76 percent in 2000 to 86 percent in 2005 but then remained virtually unchanged in 2011 at 85 percent. In rural areas the percentage increased from less than 1 percent in 2000 to 2 percent in 2005 and 5 percent in 2011.

Similar to My study the population survey of EDHS 2011 indicated that, the age structure of the household population in Ethiopia is typical of a society with a young population. The population pyramid shows the sex and age distribution of the population. The pyramidal age structure reflects the large number of children under age 15. Children under age 15 account for nearly half (47 percent) of the total population, a feature of populations with high fertility levels, while only about 4 percent of Ethiopians are over age 65. This population distribution is similar to that observed in the 2000 and 2005 surveys.

CONCLUSION AND RECOMMENDATION

Health profile assessment of the district indicates that even if there are progresses on other health related infrastructures like telecommunication, electricity power supply, drinking water, presence of all whether road but with regards to health facility shows lower coverage, health worker with in the facility showed under standard proportion to population as number of population grown rapidly.

Based on the result of an assessment current profile I strongly recommend the following list of my observation.

- Oromia Regional health bureau, Sebeta Health center, Sebeta municipality office should give special attention to provide hospital which was under construction.
- Sebeta health centers should pay special attention to promote both in of number health worker and qualification.

Since sanitation is one of the most crucial part to promote the health of the community, Sebeta town Municipality should improve waste disposal management near to public access like schools, water sources, market areas and community awareness is mandatory.

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CHAPTER 4. EVALUATION OF PUBLIC HEALTH SURVEILLANCE

4.1. Surveillance System Evaluation of Malaria, Oromia Special Zone of Sebeta Health Centers 2018.

Summary

Background: Public health Surveillance is ongoing process of gathering, analyzing, and dissemination of information for the purpose of proper planning, implementation, and evaluation of health service to reduce morbidity and mortality of the community.

Objective: To describe an existing malaria surveillance system (data collection, reporting and analysis) special zone of Sebeta health Center.

Methods: The evaluation was conducted using the Center for Disease Control guideline for evaluation for public health surveillance system. Data sources included a search of publically available literature, interviews with malaria surveillance personnel. A cross-sectional descriptive analysis was done for Malaria surveillance system from March 15- April 28, 2018.

Results: Conducted surveillance all reporting format, flow of report in all chain from health center to National level is well functioned. In case a suspected patient based on standard and community based case definition confirmation was done with Mic/RDT. There are surveillance tools, case definition and surveillance of malaria was evaluated based on the attributes.

Conclusion: Surveillance guideline for Malaria was performed in well-established way indicating to describe the trend whenever there is prevalence detected beyond the threshold the system enable to detect an outbreak easily.

Recommendation: based on conducted surveillance evaluation I strongly recommend to as surveillance guideline and performance of malaria was properly performed surveillance system and guideline for the other diseases has to be established.

4.1.1. INTRODUCTION

Surveillance is the process of gathering, analyzing, and dissemination of information for the purpose of proper planning, implementation, and evaluation of health services/interventions. It is also defined as "Information for Action". A functional disease surveillance system is essential for defining problems and taking action. Proper understanding and use of this essential epidemiological tool (public health surveillance) helps health workers at the district and health units to set priorities, plan interventions, mobilize and allocate resources, detect epidemics early, initiate prompt response to epidemics, and evaluate and monitor health interventions. It also helps to assess long term disease trends.

Malaria is estimated that three-fourths of the land below 2000 meters is malarious with two-thirds of the country's population at risk [1]. This makes malaria the number one health problem in Ethiopia with an average of 5 million cases a year and 9.5 million cases per year between 2001- and 2005[2]. The disease causes 70,000 deaths each year and accountant for 17% of outpatient visits to health institutions. It also accounts for "…15% of admissions and 29% of inpatient deaths" - a figure considered to be too low given that more than a third of the country's population does not have access to health services . A number of contributing factors have been identified. It is one of the most severe public health problems worldwide with 300 to 500 million cases and about one million deaths reported to date, 90% of which were reported from Sub Saharan African countries. It is the fourth leading cause of death of children under the age of five years in developing countries [3].

The purpose of this system evaluation is to evaluate on usefulness, reporting, data storage end results achieved as conducted surveillance of an existing Malaria surveillance which conducted in Sebeta district.

4.1.2. Objectives of Surveillance Evaluation

General Objective

To describe an existing malaria surveillance system (data collection, reporting and analysis) in special zone of Sebeta health Center.

Specific Objective

- To describe the way surveillance conducted.
- To evaluate the main important attribute of the surveillance system
- To give recommendation the way forward to improve the surveillance system whenever seemed to be improved

4.1.3. Literature Review

Several organizations or institutions have developed their own approaches for conducting evaluations of surveillance systems, and for providing relevant recommendations. These approaches path the way for developing evaluation strategies, to help evaluators to generate data in order to address specific evaluation question(s) required for management and decision-making [4].

Although the need for effective surveillance systems has long been recognized, there is increasing international pressure to improve the effectiveness of those systems even further [5]

The capacity of surveillance systems to accurately describe patterns of diseases is of public health importance. Therefore, regular and relevant evaluations of these systems are critical in order to improve their performance and efficiency [6]. Public health surveillance activities are generally authorized by legislators and carried out by public health officials. Public health surveillance systems have been developed to address a range of public health needs. In addition, public health information systems have been defined to include a variety of data sources essential to public health action and are often used for surveillance [7]

A public health surveillance system is useful if it contributes to the prevention and control of adverse health-related events, including an improved understanding of the public health implications of such events. A public health surveillance system can also be useful if it helps to determine that an adverse health-related event previously thought to be unimportant is actually important. In addition, data from a surveillance system can be useful in contributing to performance measures [8]

Public health surveillance is the ongoing, systematic collection, analysis, interpretation, and dissemination of data regarding a health-related event for use in public health action to reduce morbidity and mortality and to improve health [9]. Public health surveillance activities are generally authorized by legislators and carried out by public health officials. Public health surveillance systems have been developed to address a range of public health needs. In addition, public health information systems have been defined to include a variety of data sources essential to public health action and are often used for surveillance [10].

4.1.4. Methodology

4.1.4.1. The study area: is conducted in special zone of Sebeta Health centers which located in 25 km From Addis Ababa is bordered on North West with Walmara district, On North Burayu, on East by Akaki. Out of the total 153,612 population majority of them under 25 years, 51% and 49% of female and male consecutively.

4.1.4.2. Study Area Selection

The study area was selected purposively based on the assessment of the priority disease and up on discussion with district PHEM representative and the most prevalent results of which daily and

weekly reportable disease based on the Ethiopian Public Health Emergence management (PHEM) guide line.

4.1.4.3. Study Period: Across-sectional study method was applied from March 15-30,2018

4.1.4.4. Data Collection

Based on the guideline of CDC updated on evaluation of the public health surveillance system, on each component an interview was made with district health center PHEM officer and malaria focal person. The most activities included in interview about the case definition, the way to data collection, interpretation of the case result, analysis and reporting system and important attributes both in qualitative(usefulness, simplicity, flexibility, and acceptability) and quantitative (Sensitivity, predictive positive value, representativeness and Timeliness) was performed.

4.1.4.5. Data Analysis: Data was collected, entered and analyzed in MS Excel software Application.

4.1.5. Case definition

A case definition is according to Ethiopian Public Health Emergency management system(PHEM) guideline, 2012 a set of criteria used to decide if a person has a particular disease, or if the case can be considered for reporting and investigation as classified in to two group Community and standard case definition.

Standard case definition: is a case definition that is agreed upon to be used by everyone within the country. It can be classified as confirmed, probable, and possible or suspected. A standard case definition of suspected and confirmed cases of the 20 reportable diseases These definitions must be used at all levels including the community, health professionals working at health posts, health centers, hospitals, health offices at different levels, private health facilities, other government health facilities and NGO clinics.

Community based case definition: Is a case definition of disease and conditions adapted to suit to health extension workers (HEWs) and community members including community health workers, traditional healers, birth attendants, Kebele administration, agricultural workers,

teachers, drug outlets, etc. The community case definitions were modified for simplicity and ease understanding by HEWs and the community members.

It is clear that surveillance could not be carried out for all diseases and conditions. Therefore, priority should be given to those diseases that are of interest at national and international levels. In Ethiopia 20 diseases (13 are immediately reportable whereas 7 are weekly reportable) are selected to be included into the routine surveillance. Malaria is out of priority disease categorized in weekly reportable disease having the following standard and community based case definitions.

Standard case definition:

Suspected: Any person with fever or fever with headache, rigor, back pain, chills, sweats, myalgia, nausea, and vomiting diagnosed clinically as malaria.

Confirmed: A suspected case confirmed by microscopy or RDT for plasmodium parasites.

Community based case definition: Any person with fever OR fever with headache, back pain, chills, vomiting OR suspected case confirmed by RDT.

4.1.7. Results

Malaria is one of the priority disease of weekly reportable based on the criteria of the public health importance, mortality, morbidity (Attack rate) as it is endemic to Ethiopia, the reporting flow from Government health centers: Alemgena, Daleti and wolette and private specialty clinic, specialty center medium clinic of confirmed and species identified was sent to special zone Sebeta Health center PHEM officer and malaria focal person. Every Monday Morning in hard copy, Sebeta Health center PHEM officer and Malaria focal person received report, enter the data in to MS Excel and compile till Tuesday and sent to National Public Health Emergence management (PHEM). Following the flowchart of the PHEM guideline as illustrated in diagram *32*.

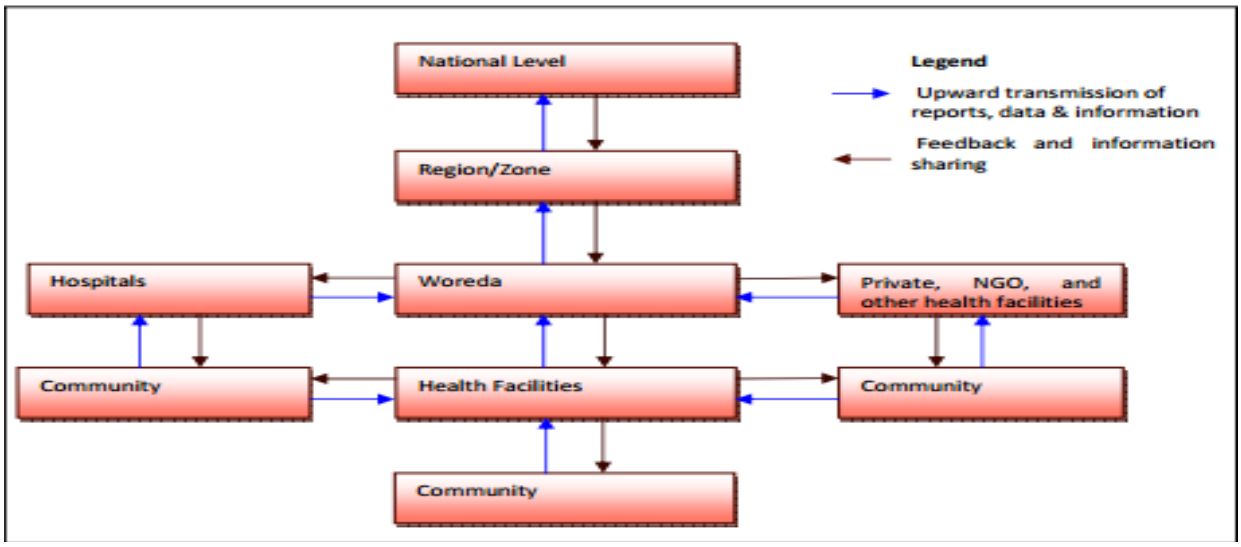

Figure 31: Illustrating the Formal and informal flow of surveillance data and information through health facilities (source, Ethiopian PHEM, Guideline 2012)

Trend of Malaria for five years from 2013-2017 indicates that the highest confirmed in 2017, 1208, the least trend in 2013(322). In 2017 there are 1208 confirmed case was reported with regards to seasonal pattern as indicated for 12 week indicated in Figure 33.

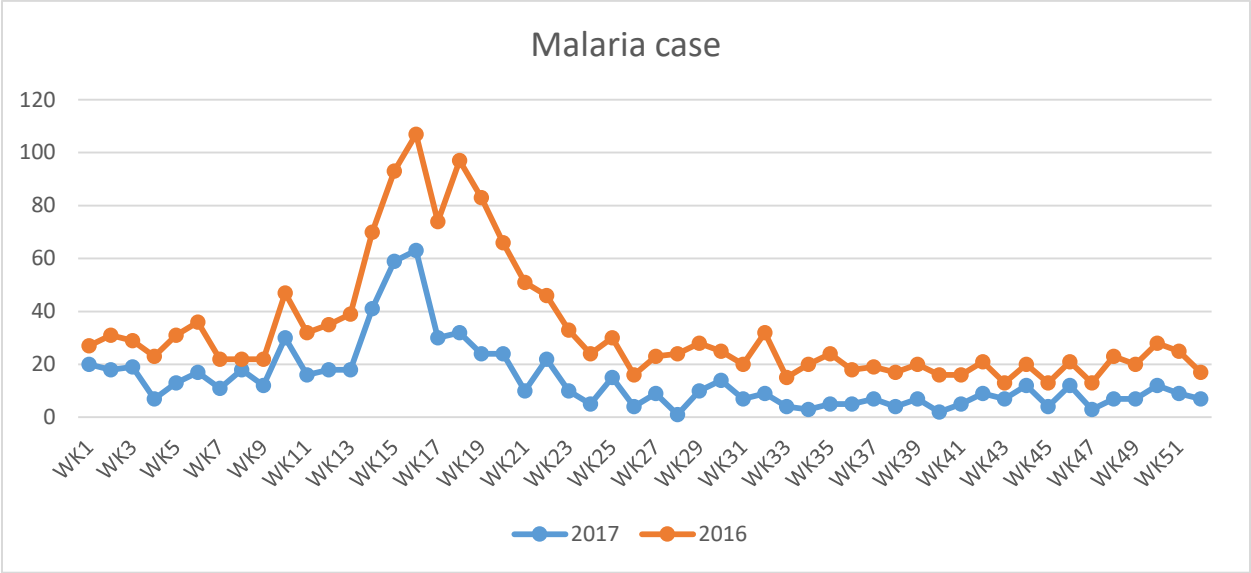

Figure 32: Trend Of Malaria Confirmed Of Sebeta District Health Centers In 2016-2017.

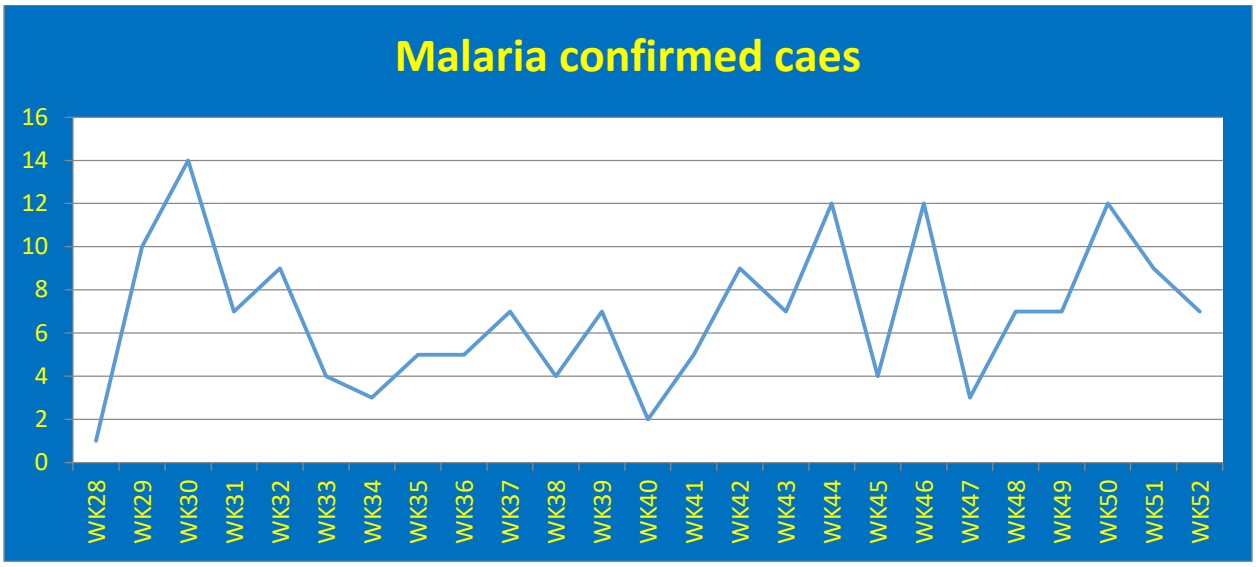

Figure 33: Figure 4. 3. Trend of Malaria in Week of Sebeta District Heath Centers in 2017.

Availability of Surveillance System Tools:

The main surveillance tools are those helps to guide to conduct surveillance, presence of case definition to define a particular disease, Reporting format according to the priority disease listed either daily or weekly reportable disease, surveillance guideline which explain the priority disease, reporting system.

Case definition: There is a standard case definition which is posted on wall visible area where everybody can access and read on each health center whereas I did not seen community based case definition in each health centers.

Reporting format: Each malaria case from Health centers and private health facilities were registered in excel sheet and stored electronically.

Evaluation based on Attributes

This section provides definitions of surveillance systems attributes based on previous guidelines for the evaluation of surveillance systems [11]. Feedback from ECDC's national surveillance coordinators who participated in a survey in 2010 [12] and working group discussions on surveillance system quality.

Simplicity: currently undergoing surveillance system in Malaria is so simple that data collection is through email, Telephone and hard copy from each government and private health facility, and data were entered in MS Excel every Monday, edited, aggregated and sent to regional, National public Health Emergence Management (PHEM).

Acceptability: based on PHEM Guideline, Report all confirmed or suspected cases of malaria on a weekly basis. If the epidemic threshold is surpassed then start reporting on daily basis using the Daily Epidemic Reporting Format for Woreda (DERF-W) and Daily Epidemic Reporting Format for Regions (DERF-R). Every collected data from health centers, private clinics sent to District Health center every Monday according to the format provided to each facility timely and all important variables.

Sensitivity: Based on the surveillance result the ability to detect an outbreak if the number of confirmed cases beyond endemic threshold health centers made regular malaria trend and evaluate each diseases with district PHEM officer and Malaria focal person.

Predictive positive value (PVP): is the proportion of all clinically suspected cases in comparison with confirmed in wet film microscopic examination. Out of suspected 20,356 cases confirmed with MIC/RDT was 9.5%.

Representativeness: to determine the representativeness of the sampling surveillance all suspected malaria case was sampled and examined MIC/ RDT was recorded in each recording case registration book and every weekly aggregated data was sent to district Public Health Emergence Management (PHEM).

Cost: with regarding cost to perform surveillance is not allocated for malaria surveillance since surveillance system is performed data source in case the trend and some indicators showed pandemic situation possible to use the budget from other source.

Supervision: from all health facilities for the surveillance of Malaria and other cases, there is no any supervision.

Prepared plan for the surveillance, there is no any plan to evaluate the achievement of the malaria surveillance

Table 17: Surveillance data evaluation performance of Malaria in Sebeta District based on the attributes, 2018.

4.1.8. Discussion

Evaluation based on the attributes indicated that acceptability, flexibility, and usefulness and

S/N	Evaluation Variables	performance in (%)	Explanation
1	Acceptability	100%	all confirmed case
2	Flexibility	100%	
3	Completeness	85%	Missing data
4	Timeliness	90%	delay one day
5	Representativeness	95%	All reported
6	Usefulness	100%	use for action
7	Predictive positive value	90%	could detect cases
8	Simplicity	100%	

simplicity performed 100% during our evaluation and interview of the health center workers, completeness there were some private health facility missed 15 % of the surveillance data which lowered to 85%, Timeliness as indicated in the PHEM reporting flow chart there were some delay of arrival of the weekly report for the Woreda PHEM office to compile the report.

Usefulness implies that surveillance results are used for public health action. Assessing usefulness consists in taking inventory of actions that have been taken in conjunction with the surveillance system. A public health surveillance system is useful if it contributes to the prevention and control of adverse health-related events, including an improved understanding of the public health implications of such events. A public health surveillance system can also be useful if it helps to determine that an adverse health-related event previously thought to be irrelevant is actually important. In addition, data from a surveillance system can be useful in contributing to performance measures, including health indicators that are used in needs assessments and accountability systems [13].

The WHO has suggested potential epidemiologic milestones for these transitions [14]. The move from a control program to a pre-elimination program involves the demonstration of a blood slide positivity rate (SPR) of less than 5% for suspected malaria cases. Subsequent transition to the

elimination phase requires reaching an annual parasite index (API) of less than one case per thousand population at risk per year [15].

Greatly intensified efforts to control and prevent malaria on a global scale have resulted in substantial reductions in malaria associated morbidity and mortality, with global malaria deaths decreasing by 47%, and incidence by 30% from 2000 to 2013, according to World Health Organization (WHO) estimates [16]. Aggressive control efforts have permitted many countries to begin the transition to elimination activities [17] which specifically seek to interrupt local transmission. With increasingly finer focus, programs concentrate their attentions from the national scale to the residual foci of active transmission, and finally to individual cases [18].

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CHAPTRE 5: PROJECT PROPOSAL

5.1. Detection and Molecular Epidemiological Study of Animal and Human Brucellosis in Eastern Ethiopia

5.1.1. Introduction

Brucellosis is a Zoonotic disease that leads to considerable morbidity resulting in significant loss of working days across the globe and thus perpetuates poverty. The disease is presented as an acute or persistent febrile illness with a diversity of clinical manifestations [1]

The disease occurs worldwide, except in those countries where bovine brucellosis *(B.abortus*) has been eradicated. The disease remains endemic among Mediterranean countries of Europe, Northern and Eastern Africa, Near East countries, India, Central Asia, Mexico, Central and South America. Although *B. melitensis* has never been detected in some countries, there are no reliable reports that it has ever been eradicated from small ruminants [2]. Furthermore, brucellosis is also considered as a re-emerging problem in many countries such as Israel, Kuwait, Saudi Arabia, Brazil and Colombia, where there is an increasing incidence of *B. melitensis* or *B. suis* biovar infection in cattle [3].

Ethiopia, located in Eastern Africa, is predominantly an agrarian country with over 85% of its population engaged in agricultural activity. The country has diverse agro ecological zones, which have contributed to the evolution of different agricultural production systems. Animal husbandry forms an integral part of agricultural production in almost all ecological zones of the country [4]. Brucellosis prevalence studies have been conducted in different localities of the country. Even then, there is little information on specific transmission dynamics within Ethiopia. The first published cases of the disease date back to 1970s [5]. This study is to attest the missing datum from an areas where people living closer to livestock and consume raw milk as a primary feed.

5.1.2. Literature Review

The genus Brucella are Gram-negative, facultative, intracellular coco-bacillary comprised of species based upon biochemical features and their correlation with preferred host species [6]. This has been an area of controversy, with species status being revoked on the basis of 16S rRNA gene sequencing, however subsequently, this decision was revoked [7].

Currently ten species are recognized including the better known six classical species comprised of *B.abortus*(cattle, biovars 1-6, and 9), *B.melitensis* (goats, sheep, biovars 1-3), *B. suis* (pigs, reindeer and hares, biovars 1-5), *B. ovis* (sheep*), B. canis* (dogs) and *B. neotomae* (desert wood rats). More recently, new members to the genus include *B. ceti* and *B. pinnipedialis* (dolphins/porpoises and seals respectively), *B. microti* (voles) and *B. inopinata* (reservoir undetermined) [8].

The incubation period varies between 14 and 120 days [9]. Primary clinical manifestations of brucellosis among livestock are related to the reproductive tract. In highly susceptible non-vaccinated pregnant cattle, abortion after the 5th month of pregnancy is cardinal feature of the disease[10]. Retention of placenta and metritis are common sequels to abortion[11]. Females usually abort only once, presumably due to acquired immunity. In general, abortion with retention of the placenta and the resultant metritis may cause prolonged calving interval and permanent infertility. In humans, the disease is characterized by a multitude of somatic complaints, such as fever, sweating, anorexia, malaise, weight loss, depression, headache and joint pains and is easily confused with malaria and influenza[12].

Various synonyms have been used for human brucellosis including Malta fever, Rock fever of Gibraltar, Cyprus or Mediterranean fever, intermittent typhoid and most frequently, undulant fever [13].

Statement of the problem

Brucellosis primarily affects livestock, but can be transmitted to humans by consumption of raw milk, close contact with aborted fetus, inhalation or accidental inoculation of suspected samples. The prevalence of human brucellosis differs between areas and has been reported to vary with standards of personal and environmental hygiene, animal husbandry practices, and species of the causative agent and local methods of food processing[14].

According to the Brucellosis 2003 International Research Conference estimated that 500,000 human infections occur per year worldwide, with incidences ranging from less than one case per 100,000 population in UK, USA and Australia, through 20 to 30 cases per 100,000 in southern European countries such as Greece and Spain, to more than 70 cases per 100,000 in Middle Eastern States such as Kuwait and Saudi Arabia [15].

As compared to study of animal brucellosis, study of human brucellosis in Ethiopia is sparse with even less information on risk factors for human infection. For instance, out of 56 cases with fever of unknown origin, two (3.6%) were reported to be positive for *B. abortus* antibodies by RBPT and CFT [16]. A study conducted in traditional pastoral communities by [17] using *B. abortus* antigen revealed that 34.1% patients with febrile illness from Borena, 29.4% patients from Hammer and 3% patients from Metema areas were tested positive using Brucella IgM/IgG Lateral Flow Assay. But they failed to include a parallel study of animal brucellosis. Studies conducted in high risk group such as farmers, animal health professionals, meat inspectors and artificial insemination technicians in Amhara Regional State [18].

5.1.3. Objective

General Objectives

To estimate Sero-prevalence, molecular detection and characterization of brucellosis in cattle, small ruminant, camel and human.

Specific objectives

- To estimate the prevalence of both animal and human brucellosis in Eastern part of Ethiopia
- To characterization the laboratory confirmed cases
- To perform phylogenies to determine the present species of country

5.1.4. Method and material

4.1.4.1. Study area

The study will be conducted at Eastern and West part of Ethiopia, Oromia region(West Hararge and Horo Guduru Wollega), Somali region (Fafen and Sitti) districts, Benishangul Gumuz region(Asosa and Metekel) zones (Kamashi and Wanbara districts) of randomly selected four kebeles from each districts.

Sampling technique: Whole blood will be collected with Vacationer tube with heparinized tube and milk sample from animal with abortion history, both serum and clot blood were kept with the same id and kept in refrigerator and transported to National laboratories (NAHDIC and EPHI)

Sample size determination based on the prevalence of the previously studied which is 4-8% about 800 samples will be collected from animal and human.

sampling Size: calculated based on single proportion calculation $n=(z^2)P(P-1)/d2$ =369 this means based on the sample size determination prevalence bases is 369 from this total sample size 147.6(40%) cattle, 129(35%) camel and 92.25(25%) is small ruminants and 370 to 400 blood samples from human where there is an animal sample were taken.

5.1.4.2. Sampling Methods:

The study Method will be cross-sectional study one time blood sample taken from animal and human perform an individual test first serum in ELISA those positive in serology will go for Molecular antigen detection for confirmatory both Genus and species for *B.abortus* and *B.melitensis*. PCR positive samples will be sent for sequences either capillary based or NGS platform data will be interpreted in different bioinformatics tool to perform phylogenetic tree for molecular epidemiology.

5.1.4.3. Data Analysis

The result of the study data analysis will be performed with SPSS and epi info 7.1 software and different tools of bioinformatics and interpreted the molecular epidemiology.

Ethical consideration

Ethical clearance to conduct the study were certified from Addis Ababa University School of public health Ethical review board.

Dissemination of information

The result of the study will be communicated with Addis Ababa University School of Public health Department of field epidemiology

National Animal Health Diagnostic Investigation Center (NAHDIC),

Ethiopia Public Health Institute (EPHI)

Expected outcomes:

There will be clear prevalence data will be available as national wise

There will be clear epidemiological linkage between animal and human as a result of close contact as source of exposure of aborted fetus, consumption of raw milk.

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.CHAPTER 6. SCIENTIFIC MANUSCRIPT FOR REVIEW

6.1. Detection of Anthrax from Farta and Smada Districts of South Gondor, Amhara Regional State, Ethiopia, April, 2019.

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Key Words: Anthrax, Smada and Farta, Amhara.

Abstract

Background: Anthrax is a serious zoonotic disease that can affect most mammals and several species of birds, but is particularly important in herbivores. We identify an anthrax case from two district of south Gondor zone, Amhara region from March, 13 to April 8, 2019.

Material and Methods: Descriptive study design methods was used, samples were taken and an individual sample DNA extracted from Tissue with QIAamp® DNA extraction mini kit, amplification was done with Real time PCR for confirmatory test.

Result: All samples from animal taken from March 13-April 8,2019 from Farta and Smada districts from two PA's were positive with Real time PCR, there are 17 cases from two districts 5 female and 12 male and one death of female of 45 year old were investigated. An index case was a person with 55 years old on March 13,2019 duration of an outbreak is till 8Aprill,2019, but still there is an animal case is happening because the spore were widely dispersed with in the District.

Conclusion and recommendation: All cases were confirmed of Anthrax, the two districts were an endemic area of anthrax, there was a death before two years with similar situation, since the vaccination coverage is under standard almost all livestock with in the outbreak and adjacent and human is still under risk. Awareness creation, routine vaccination of an animal is mandatory.

6. 1.1. Introduction:

Anthrax is a worldwide zoonosis that primarily affects herbivorous animals [1in domestic animals there is high fatality in cattle, sheep goats, horses, donkeys and pigs [2] for wild animals. A disease caused by the bacterium *Bacillus anthracis.* It is a gram-positive endospore forming , rod-shaped , non- motile , bacterium that grows in nutrient media, typically sheep or horse blood, under aerobic and anaerobic conditions with an optimal temperature of 35-37 ^{0} [3, 4]

Humans are known as accidental hosts and usual become infected by accidental contact with spores or vegetative bacilli either through the soil, insect or through direct contact with infected animal carcasses or material. Human at the highest risk of occupational exposure are veterinarian, livestock farmers, any personnel that handle animal product (such as butcher, wool sorter, tannery workers, etc.), and laboratory personnel [4]

Spores survive for decades in soil that is rich in calcium, has a PH greater than 6.0 and when temperatures are higher 15.5 ^{o} [5], it is considered a seasonal disease as outbreak tend to follow a prolonged hot and dry period that is followed by heavy rainfall. It is thought that heavy rain can carry spores during runoff in clumps of organic matter to concentrate in standing pools or puddles [5].

South Gondor is one of the Amhara regional state with different agro-ecological zone from 3900mal to 2278 mal, covered with high mountain area, with different districts of this zone there is long time trend that traditional Tannery was carried out. The main objective of this study was to detect Anthrax case and confirm with Real Time PCR for the first time as national wise, for appropriate outbreak response and recommendation.

6.1.2. Methodology: the study area was where an outbreak of anthrax happened in Farta and Smada districts of south Gondor zone, Wowa Magera and Dudubia (Kebele20) Kebeles.

Sampling method: based on epidemiological data from an outbreak area, Tissue sample was collected with biohazard bag triple package with appropriate decontamination with 10% bleach, kept ice box and transported to National Animal Health diagnostic and Investigation Center (NAHDIC).

6.1.3. DNA extraction:

DNA Extraction was performed from tissue sample with appropriate biosafety measure and decontamination with 10% bleach in BSL-3 zoonotic laboratory, each sample were individually extract with QIAamp DNA extraction mini kit. Tissue was rinsed with PBS to avoid inhalation of spore from dried tissue, then minced with sterile scissors and grinded with mortar and pistil with fine sterile sand, after formed suspension, Extraction started with lysis buffer added 180 micro liter of ATL buffer, then 20 µl proteinase K and vortexed. Decontaminate with 10% bleach then incubated with 56^{o} for 1-3 hours vortex every 30 minutes.

After 3 hour incubation in water bath at 56^{o,} the whole volume was filtered with Millipore to avoid spore not to be extracted with other cells, transfer to new mini-spin column added 200 µl of Al buffer, vortex then for binding added 200 µl of absolute alcohol (Ethanol) vortex centrifuge 8000rpm for 3 minutes. After binding step washed with washing buffer one (AW1) centrifuged at 8000rpm for 3 minutes, Wash two (Aw2) with similar procedure. Finally elusion was done with 200 µl of AE buffer followed by centrifuge at 8000rpm for 3 minutes, DNA was kept at -20^{0} till Amplification.

6.1.4. DNA Amplification

Bacterial amplification was performed with Real-time PCR comprised with master-mix components; primers which targeted for both protective antigen (PA) and Capsular antigen (CAP) both forward and reverse primers and probes for each. Protective antigen (PA) PA- Forward 5 ' CGG ATC AAG TAT ATG GGA ATA TAG CAA - 3' PA- Reverse 3' CCG GGT TAG TCG TTT CTA ATG GAT 5' PA-probe: FAM CTC GAA CTG GAG AGT GTT ACC GCA AAT-BHQ1

Capsular antigen (CAP) Cap-Forward: ACG TAT GGT GTT TCA AGA TTC ATG. Cap-Reverse: ATT TTC GTC TCA TTC TAC CTC ACC. Cap Probe: FAM- CCA CGG AAT ATC TCA AAT GGC AT-BHQ1.

In Master-mix component 10X Exo IPC Mix and 50X Exo IPC Mix DNA were added to monitor inhibitory substance with in Real time PCR. Thermal cycler protocol was Denaturation 95^{o} for 10 minutes, Amplification 95^{o} for 15 seconds and 60^{o} for 1 minutes for 40 cycle's standard mode of 7500 with passive reference of ROX.

6.1.5. Data Processing and Analysis: The data were entered and analyzed using Epi-Info7 version 7.1.0.6. Results were presented using graph and table. Attack rate and case fatality rate were also calculated.

6.1.6. Result

6.1.6.1. Descriptive by place:

We identified for an epidemiological association with hypothesized potential risk Factors. According to the district veterinary clinic report, Anthrax vaccination performance was 16% which is much lower than acceptance range. An anthrax case from two districts the higher case in people who were in Farta district of Wowa Magera PA with widely spread case almost in the whole Kebele.

6.1.6.2. Descriptive by person:

Since there is no anthrax vaccine in human in Ethiopian context, whenever there is an animal outbreak or people exposed to spore very prone to anthrax case, from two district and two PA's from anthrax case death of one cow 5 persons who involved in skinning and slaughtering and 4 who involved in deboning parting the organs 8 of them were develop cutaneous case , out of 46 households who exposed with meat of dead animal 9 of them develop cutaneous case and admitted to Debre-tabor hospital and 1 adult female who exposed and did not visit health facility was dead of anthrax after 24 days of dead of cattle and 15 days of index case of anthrax case, totally there are 18 cases from two districts.

Table 18: Anthrax Case Of Wowa Magera Kebele, Farta District, South Gondor Zone, Amhara Regional State April, 2019

Wowa Magera	population	Case	Percentage	AR/1000 population	Death	CFR
Male	2678	12	70.59	4.5	0	0
Female	4658	5	29.41	1.10	1	20
Total	7336	17	100	2.32	1	5.9

Table 19: Anthrax Case of Dudubia Kebele, Smada District, South Gondor Zone, Amhara Region, April, 2019.

Dudubia	**population**	**case**	**Percentage**	**AR/1000 Population**	**Death**	**CFR**
Male	2003	1	100	0.5	0	0
Female	2006	0	0	0	0	0
Total	4009	1	100	0.25	0	0

Assessment on cutaneous anthrax based on our questioner survey sign and symptom of the onset of the disease 95% of them develop swelling with CI of 95% as a result of statistical data showed in Table 3.3. And 88.24 % seen itching with Level of Confidence 95% as indicated in table 18.

Table 20: Frequency of Respondent's Anthrax Case Swelling Sign, Wowa Magera Kebele, Farta District, South Gondor Zone, Amhara Region, and April, 2019

Swelling response	Frequency	Percent	Cum. Percent	Exact 95% LCL	Exact 95% UCL
Yes	16	94.12%	94.12%	71.31%	99.85%
No	1	5.88%	100.00%	0.15%	28.69%
TOTAL	17	100.00%	100.00%		

Table 21: Frequency Of Respondent's Anthrax Case of Itching Sign, Wowa Magera Kebele, Farta District, South Gondor Zone, Amhara Region, 2019.

Itching	Frequency	Percent	Cum. Percent	Exact 95% LCL	Exact 95% UCL
Yes	15	88.24%	88.24%	63.56%	98.54%
No	2	11.76%	100.00%	1.46%	36.44%
TOTAL	17	100.00%	100.00%		

6.1.6.3. Descriptive by Time

The onset of an outbreak is after 5 days post exposure of dead animal carcass which is a cow dead on 14 of March 2019, first case was on 19 of March,2019 three cases and visit to hospital followed by others with different date till 3rd of April 2019, as others discharged from hospital in average of 10-12 days of inpatient, one serious case from Dudubia of Smada district admitted on March 27 to April 8 still in hospital for 12 days not yet discharged till our last visit on 8th of April,2019.

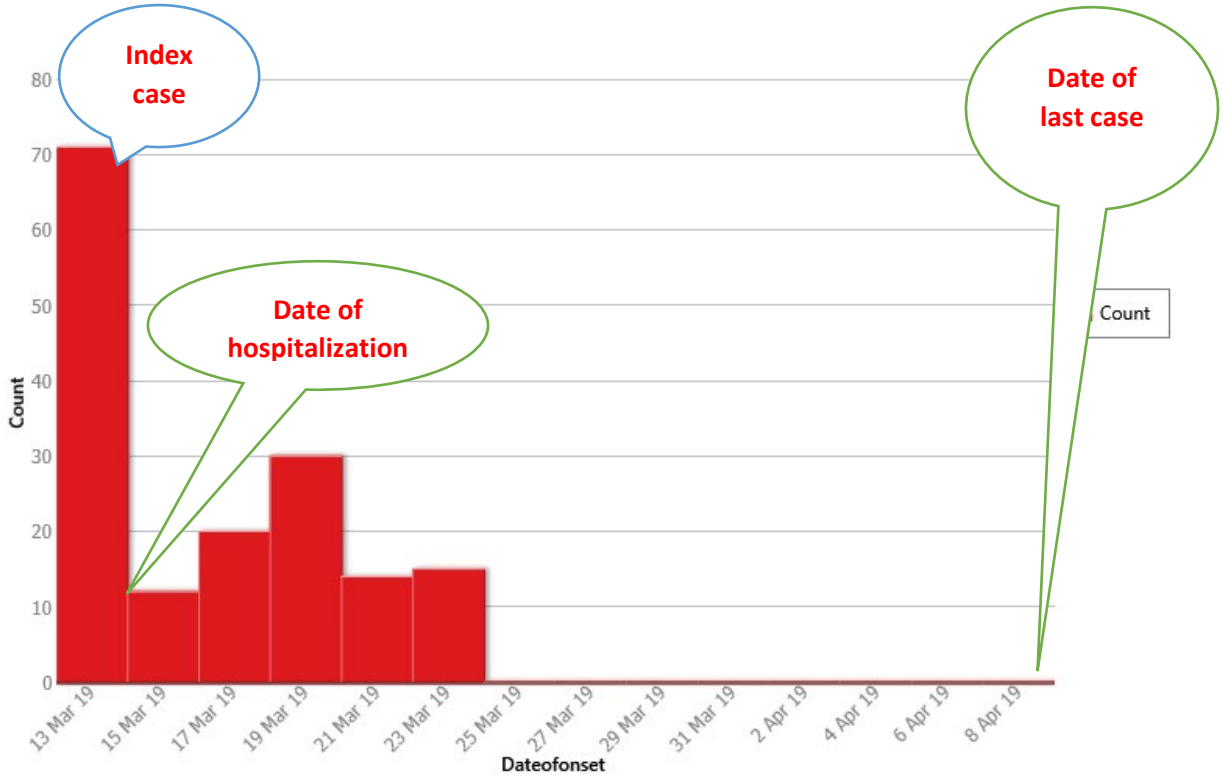

Figure 34: Epidemic Curve of Anthrax Outbreak in Farta and Smada Districts of South Gondor, From March-April, 2019.

6.1.6.4. Laboratory Result:

All cases of animal dead sampled were examined and confirmed with PT-PCR, at National Animal Health Diagnostic and Investigation Center (NAHDIC) In Molecular Diagnostic laboratory. This is the first time that an anthrax case confirmed in Ethiopia as national wise with Real time PCR. As some literature documented there are about 1200 different strains of Anthrax were available in worldwide still no one knows Ethiopian strain, those sample who were positive with Real time

PCR was stored in -20º both DNA and crude samples for further characterization to identify which strain is circulating in our country.

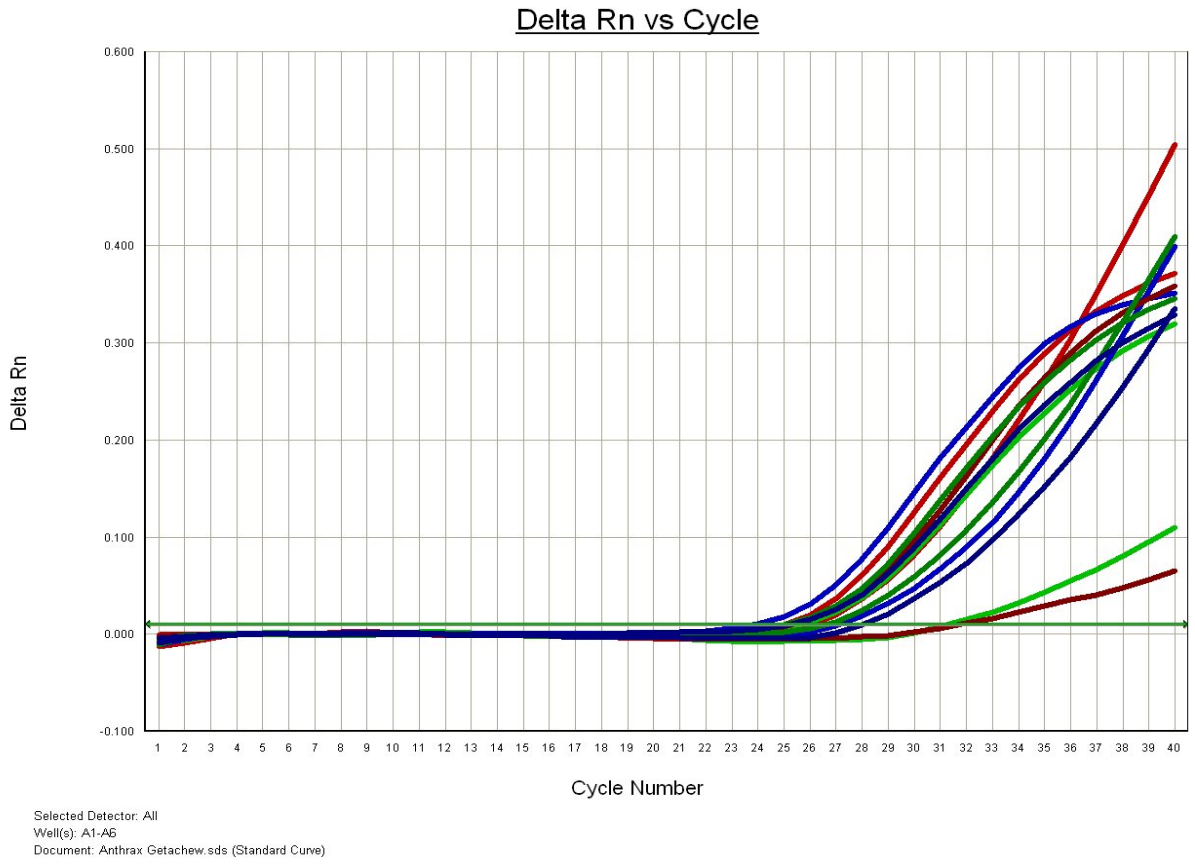

Figure 35: Real Time PCR Result of Amplified B.anthracis Bacteria With Applied Bio-Systems 7500 At National Animal Health Diagnostic And Investigation Center (NAHDIC) April, 2019.

6.1.7. Discussion

Even though there are several suspected anthrax cases with in the country both from animal and human from year to year, this investigation followed all epidemiological investigation suspected animal sample was taken and confirmed with real time PCR, and this is the first time that confirmed anthrax from Ethiopia with Real time PCR.

A century's experience of the natural disease in humans suggests that it probably does not take many spores to initiate a cutaneous infection once the necessary access to sub epidermal tissues is achieved, despite the implication otherwise in the experimental data. It is generally accepted that *B. anthracis* is non-invasive and that the spores must gain access to sub epidermal tissue through

a cut or abrasion before infection can occur, although this has been challenged: in a series of laboratory-acquired cutaneous anthrax cases, preceding trauma at the site of the lesion was noted by only 5 of the 25 patients [6].

Anthrax eschars are generally seen on exposed unprotected regions of the body, mostly on the face, neck, hands and wrists. Generally cutaneous lesions are single, but sometimes two or more lesions are present. For example, with infection resulting from skinning an infected dead animal, multiple lesions may be seen on hands, wrists or arms. The incubation period ranges from as little as 9 hours to 3 weeks, mostly 2 to 6 or 7 days. In the October–November 2001 anthrax letter cases in the USA, the mean/median incubation period for the 11 cutaneous cases was 5–6 days, with a range of 1–12 days [7-9].

Conclusion and Recommendation

This outbreak investigation was the one which confirm Anthrax for first time in Ethiopia with real time PCR test method. All cases were confirmed cases, with regards to the risk factor as South Gondor is Endemic area for anthrax, there is spore in the soil, dead animal hide and skin, feed and from the environment. As a vaccination coverage is the lowest status it play a vital role for the occurrence of the outbreak.

Based on our finding and assessment of the area the following points were forwarded to the health professional, community and administrative officials. An animal with sudden death with prognostic sign should not be opened, buried with intact skin and hide in deep with clay soil to avoid exposure of scavenger and drainage to rivers and streams. As an area and adjacent district were under risk an animal should regularly vaccinated every six month since source of infection is an animal.

There should be routine awareness creation for the community for better control of the disease in each level. We recommend also for health facilities to think about the human vaccine for further prevention of the public health.

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CHAPTER 7: SERO-PREVALENCE STUDY OF BRUCELLOSIS.

7.1. Prevalence Study And Molecular Epidemiology of Brucellosis in Eastern Ethiopia

Abstract:

Introduction: The main objective of this study is to determine the seroprevalence and identification of the causative agent, in Cattle, Small ruminant and camel from Eastern part of Ethiopia.

Methodology: A total of 698 blood samples were collected from cattle, Sheep goats and camel. Data related to age, sex, location, and breed were collected on the sampling day. Serum samples was initially screened using C-ELISA. Seropositive samples were subjected to bacterial PCR analysis using Brucella species-specific (IS711 for *Brucella abortus* and *Brucella melitensis*) real-time polymerase chain reactions (RT-PCR).

Results: Forty three (6.1%) serum samples were positive C-ELISA. Of the 43 seropositive serum samples, 20 (46.5%) were positive in the *Brucella abortus*-specific (IS711) RT-PCR. Of which 19(95%) were *B.melitensis* from all samples, only 1(5%) was *B.abortus* (5%).

Conclusions: *Brucella melitensis* and *Brucella abortus* were identified as causative agent of Camel, *B.melitensis* was for the causative agent of ovine and caprine brucellosis in the study area. Based on the study result screening of brucellosis should be practiced before an introduction of animal for breeding purpose and used as consumption of milk for human.

7.1.1. Introduction

Brucellosis is a Zoonotic disease that leads to considerable morbidity resulting in significant loss of working days across the globe and thus perpetuates poverty. The disease is presented as an acute or persistent febrile illness with a diversity of clinical manifestations [1].

The disease occurs worldwide, except in those countries where bovine brucellosis *(B.abortus*) has been eradicated. The disease remains endemic among Mediterranean countries of Europe, Northern and Eastern Africa, Near East countries, India, Central Asia, Mexico, Central and South America. Although *B. melitensis* has never been detected in some countries, there are no reliable reports that it has ever been eradicated from small ruminants [2]. Furthermore, brucellosis is also considered as a re-emerging problem in many countries such as Israel, Kuwait, Saudi Arabia, Brazil and Colombia, where there is an increasing incidence of *B. melitensis* or *B. suis* biovars infection in cattle [3].

Ethiopia, located in Eastern Africa, is predominantly an agrarian country with over 85% of its population engaged in agricultural activity. The country has diverse agro ecological zones, which have contributed to the evolution of different agricultural production systems. Animal husbandry forms an integral part of agricultural production in almost all ecological zones of the country [4]

Brucellosis prevalence studies have been conducted in different localities of the country. Even then, there is little information on specific transmission dynamics within Ethiopia. The first published cases of the disease date back to 1970s [5]. This study is to attest the missing datum from an areas where people living closer to livestock and consume raw milk as a primary feed.

7.1.2. Literature Review

The genus Brucella are Gram-negative, facultative, intracellular Coco-bacillary comprised of species based upon biochemical features and their correlation with preferred host species[6]. This has been an area of controversy, with species status being revoked on the basis of 16S rRNA gene sequencing, however subsequently, this decision was revoked [7]

Currently ten species are recognized including the better known six classical species comprised of *B.abortus*(cattle, biovars 1-6, and 9), *B.melitensis* (goats, sheep, biovars 1-3), *B. suis* (pigs, reindeer and hares, biovars 1-5), *B. ovis* (sheep*), B. canis* (dogs) and *B. neotomae* (desert wood rats). More recently, new members to the genus include *B. ceti* and *B. pinnipedialis* (dolphins/porpoises and seals respectively), *B.microti* (voles) and *B.inopinata* (reservoir undetermined) [8]

The incubation period varies between 14 and 120 days [9]Primary clinical manifestations of brucellosis among livestock are related to the reproductive tract. In highly susceptible non-vaccinated pregnant cattle, abortion after the 5th month of pregnancy is cardinal feature of the disease [10]Retention of placenta and metritis are common sequels to abortion [11]

Females usually abort only once, presumably due to acquired immunity. In general, abortion with retention of the placenta and the resultant metritis may cause prolonged calving interval and permanent infertility. In humans, the disease is characterized by a multitude of somatic complaints, such as fever, sweating, anorexia, malaise, weight loss, depression, headache and joint pains and is easily confused with malaria and influenza [12]

Various synonyms have been used for human brucellosis including Malta fever, Rock fever of Gibraltar, Cyprus or Mediterranean fever, intermittent typhoid and most frequently, undulant fever [13]

Brucellosis primarily affects livestock, but can be transmitted to humans by ingestion, close contact, inhalation or accidental inoculation. The prevalence of human brucellosis differs between areas and has been reported to vary with standards of personal and environmental hygiene, animal husbandry practices, and species of the causative agent and local methods of food processing [14]

According to the Brucellosis 2003 International Research Conference estimated that 500,000 human infections occur per year worldwide, with incidences ranging from less than one case per 100,000 population in UK, USA and Australia, through 20 to 30 cases per 100,000 in southern European countries such as Greece and Spain, to more than 70 cases per 100,000 in Middle Eastern States such as Kuwait and Saudi Arabia [15]

As compared to study of animal brucellosis, study of human brucellosis in Ethiopia is sparse with even less information on risk factors for human infection. For instance, out of 56 cases with fever of unknown origin, two (3.6%) were reported to be positive for *B.abortus* antibodies by RBPT and CFT [16]

A study conducted in traditional pastoral communities by [17] using *B.abortus* antigen revealed that 34.1% patients with febrile illness from Borena, 29.4% patients from Hammer and 3% patients from Metema areas were tested positive using Brucella IgM/IgG Lateral Flow Assay. But they failed to include a parallel study of animal brucellosis. Studies conducted in high risk group such as farmers, veterinary professionals, meat inspectors and artificial insemination technicians in Amhara Regional State[18]

The economic losses due to bovine brucellosis arises from slaughter of cattle herds that are infected with Brucella, loss of calves dues to abortion, birth of weak calves that die soon after birth, impaired fertility, decrease milk yield, endangering animal export trading of a nation, government costs incurred for research and eradication programs [19].

Bovine Brucellosis is primarily a reproductive disease characterized by abortion last trimester or birth of unthrifty newborn in the female and orchids and epididymitis with frequent sterility in male [20] means of transmission in both female and male is through ingestion and direct or indirect contact, environment contamination by fetus, fetal membrane vaginal secretion like pasture forage, water, stable etc. while the means by which man get infected are through indirect contact of materials contaminated by fetus, fetal membrane, vaginal secretion and directly through ingestion of raw milk and fresh cheeses (PAHD-WHD,2001)

Besides milk, meat is one of the most important products from camels. Yield and quality of the carcasses are comparable to other livestock. However, camels are still not systemically bred for meat production as they are considered too valuable for this production type. Another important

product is camel hair. It is one of the world's most expensive natural animal fibers. Camelids are appreciate for the high quality of their wool (alpaca and Bactrian camel). Recently, the camel leather industry was better valorized in relationship with the touristic activities. Camel hides are known for their strength and durability. They are used by camel breeders, but also for fashion accessories [21]

Human infection due to Brucella from camels is known to occur mostly through the consumption of raw milk [22]Malta fever due to *B.melitensis* was diagnosed in 30% of the camel handlers and milkers on large camel farm in Riyadh, Saudi Arabia. The abortion rate in the farm reaching 12% and *B.melitensis* biovars 1, 2 and 3 were isolated from aborted camel fetuses [23] Therefore, there is a real need for cooperation between public health officials and veterinary officers to reduce the circulation of human brucellosis in endemic areas [24]

7.1.3. The Study

7.1.3.1. Material and Methods

7.1.3.2. Study area

The study area was conducted in Eastern part of Ethiopia, Oromia region(West Hararge(Gemechis, Gumbi Bordode, Ciro zuria and Anchar districts , Somali region (Jigjiga) zones(Shebele district) of different Kebeles who were randomly selected in two types of agricultural practice of the study areas agricultural and pastoralists

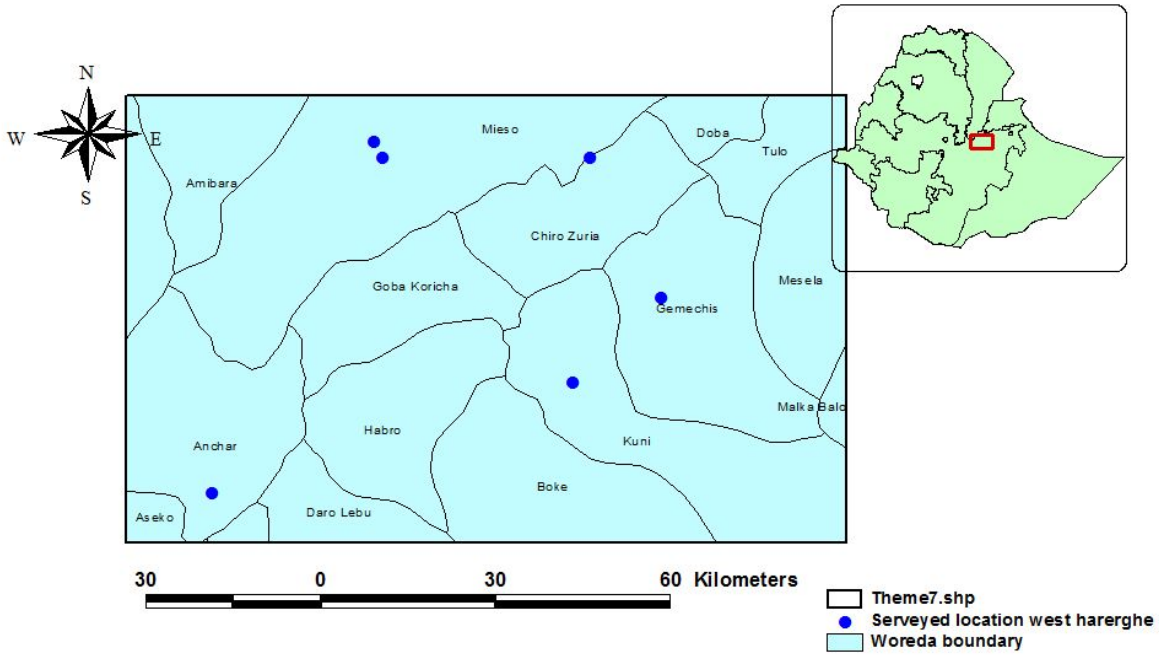

Figure 36*:* Sampling Areas, Eastern Part of Ethiopia, East Oromia and Somali Regional States, December, 2018.

7.1.3.4. Sampling technique: an individual animal Whole blood was taken from jugular vein with Vacationer tube with heparinized tube from randomly selected and milk sample from previously abortion history or related herds which has close contact with Bull that previously had abortion after mating with bulls from the herd.

7.1.4.5. Serological study

Totally there are 698 blood sample were collected from cattle , small ruminant(sheep and Goats) and Camel in 3 districts from west Hararge(Anchar, Gumbi bordode , Gemechis) and Shebelle district from Somali regional state. Serum sample were tested in C-ELISA for the detection of antibody against Brucellosis.

7.1.4.6. Molecular study

7.1.4.6.1. DNA Extraction

An individual blood clot of previously known to be positive with C-ELISA, DNA extracted 1.5 gm of blood clot was taken and added to 20microliter of proteinase K and 180 AL buffer and kept in 56^0c of water bath for one and half an hour vortex mixing every 30 minute for lysis of cells, after lysis 200 micro liter and 200 micro liter of ATL buffer was added and thoroughly mixed. Transferring the whole volume with mini spin column two step washing with AW1 and AW2 washing buffer and Eluted with AE buffer in 100 microliter of volume.

7.1.4.6.2. DNA Amplification

An extracted DNA was amplified with RT-PCR 7500 standard mode in specific primers 16S target gene first IS711 Genus(5'-GCTTGAAGCTTGCGGACAG-3') forward, (5'-CCTACCGCTGCGAAT-3') reverse and FAM-AAGCCAACACCCGGCCATTATGGT-BHQ-1probe with Internal positive control (IPC) 10x Exo IPC Mix and 50x Exo IPC DNA added after being amplified screening Brucella genus, amplification was performed for specific species primers for *B.abortus* (5'-GCACACTCACCTTCCACAACAA-3')forward, (5'-CCCGTTCTGCACCAGACT-3')reverse and *B.melitensis* (5'-TCGCATCGGCAGTTTCAA-3') forward and (5'-CCAGCTTTTGGCCTTTTCC-3') were amplified.

7.1.4.6.3. Data Analysis

The result of the study data analysis was performed with SPSS and epi info 7.1 software and different tools of bioinformatics and interpreted the molecular epidemiology.

7.1.5. Results

7.1.5.1. Serological Result Analysis

Out of 698 Blood samples from two regions and five districts of each serum tested with C-ELIZA 43(6.2%) were positive out of each 21(10 %) were from camel, 11(6.1%) from caprine and 11(19.6%) from ovine were positive, from cattle sample none of the samples were detected. As indicated in table 22.

Table 22: Serological Study Result on Brucellosis from Eastern Part of Ethiopia, March, 2019.

Sero- Prevalence in Species			
Species	**No of Testes**	**C-ELIZA Positive**	**Prevalence In (%)**
Cattle	250	0	0
Camel	211	21	10
Caprine	181	11	6.1
Ovine	56	11	19.64

As indicated from table 22. There is highest prevalence were seen in ovine followed by camel while no antibody was detected from cattle in our study.

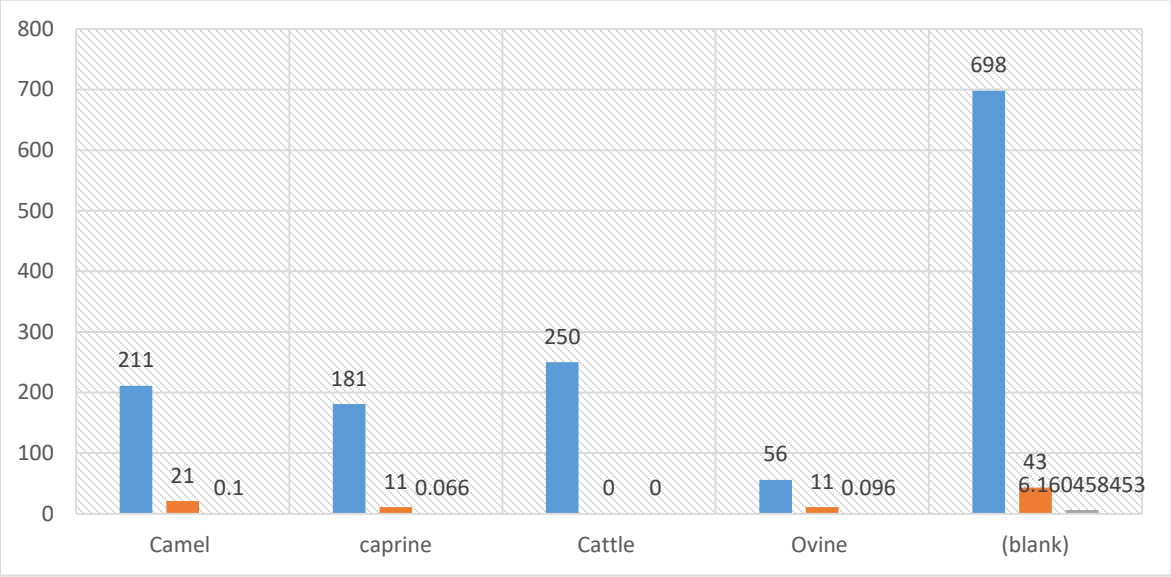

Figure 37: Serological Prevalence of Brucellosis in Eastern Part of Ethiopia, March, 2019.

Age Related Prevalence:

Serological result with age related brucellosis prevalence, there are two age group in our sampling which is young from early young up to 1 years of age and adult who were above 1.5 year old including Female adults who were on lactating, pregnant and Adult Male, our study result indicates that out of 43 sera positive with C-ELIZA 37(86.05%) were adult age group in all animal species. Indicated in table 21.

Table 23: Age Wise Distribution of Brucella Sero- Positive Animals from Eastern Part of Ethiopia, March, 2019.

Age	No. of positive	In Percent
Young ≤1 year	6	13.95(%)
Adult > 1 year	37	86.05(%)
Total	43	100(%)

Sex related prevalence

Out of collected 698 sera sample tested for antibody with C-ELIZA in both sexes to indicate as there is sex linked prevalence, 41(95.35%) were Female in all animal species which is only 2(4.65), less than 5% of them were from male samples were positive.

Table 24: Distribution of Sero-Positive Animals with Sex Related, March 2019

Sex	No. of sera positive	Percentage (%)
Male	2	4.65
Female	41	95.35
Total	43	100

An overall brucellosis study from 698 sera sample taken to determine the seroprevalence 43/698 which is 6.2% of prevalence, with regards to species prevalence Cattle no antibody was detected from 240 sera sample, Camel 21/211 with 10% Sero prevalence, Caprine 11/181, 6.1% and ovine 11/56, 19.6% of prevalence.

Molecular result Analysis

Out of 43 previously known positive with serological test performed with Real time PCR for confirmatory and species identification, 20 samples (46.5%) were positive with *IS711* Brucella Genus screening test with Ct value range between 16 and 36, With regards to animal species tested out of examined 21 camel sample 9(43%), Caprine out of 11 sample 8(73%) and ovine out 11 samples 3(27.3%) were positive with RT-PCR test methods. With regards to Brucella species identified with specific primers out of 20 positive samples with RT-PCR 19 (95%) were *B.melitensis* with only 1(5%) was *B. abortus* which is from Camel origin.

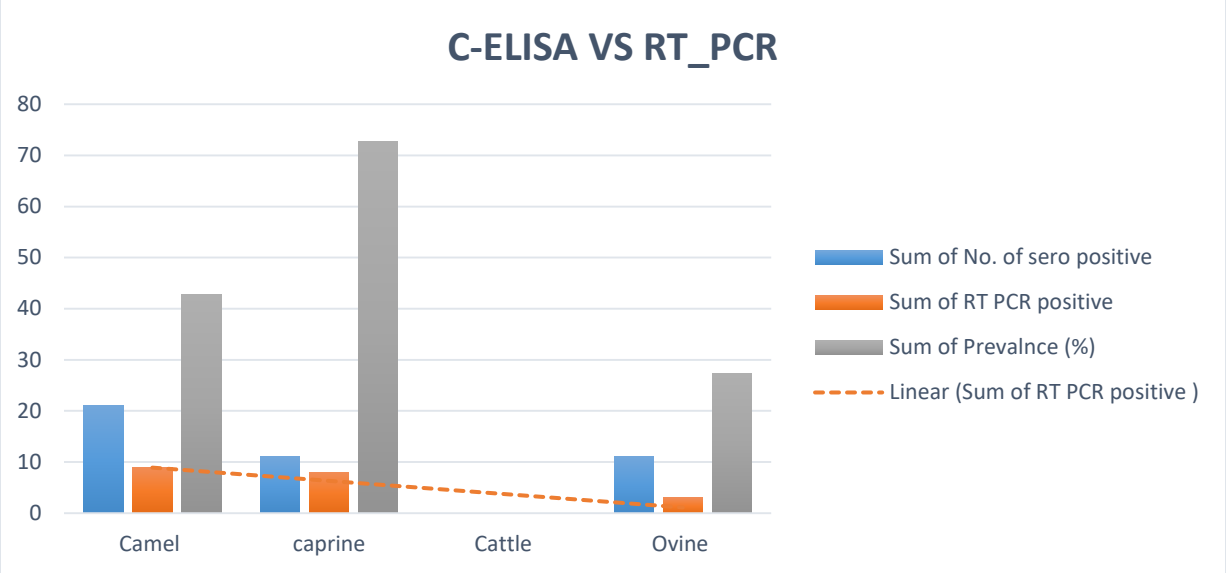

Figure 38: Prevalence of Brucellosis test result in C-Elisa with RT-PCR from Eastern Part of Ethiopia, March, 2019

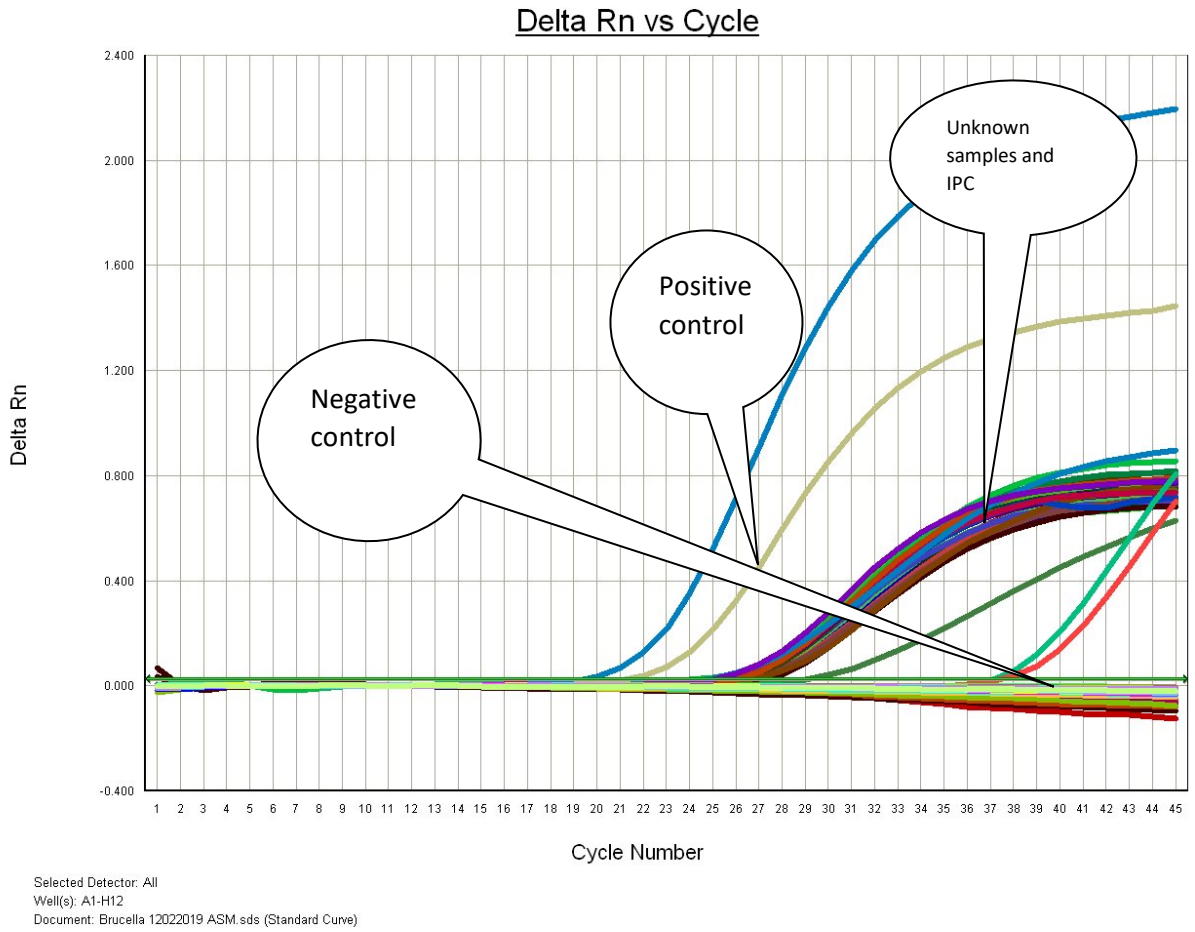

Figure 39: Amplification curve of RT-PCR result of brucellosis, April, 2019

7.1.6. Discussion:

The study showed Sero-prevalence of an animal indicated that with varied prevalence of positivity, as indicated in the result absence of Sero-positive in cattle does not mean that the animal is negative since there were abortion history during the surveillance and sampling. Both species of *B.abortus* and *B.melitensis* were detected from camel with specific primers from clot blood extracted DNA.

In comparison among the animal species camel has the highest Sero-prevalence the reason is they are so close contact with in each other, at the same time out of sampled area those animal having the same herd showed similar pattern of positivity because of contagious nature of the bacteria.

Brucellosis is a serious zoonotic disease affecting man and all domestic animals including camels. It is considered as one of the great public health problem all over the world [25]. However, camel brucellosis has not received proper attention from researchers and scientists. Brucellosis was

reported in camels as early as 1931 [26]; since then, the disease has been reported from all camel-keeping countries. Sero-prevalence of the previous study according to [27] of camel from Ethiopia was 5.5% with RBT screening test which is significantly less than our study which is 21/211 which is 10%, this may showed that the prevalence was increasing from time to time since there is no appropriate control measure was taken.

Caprine prevalence from previous Seroprevalence according to [28] article review of seroprevalence in caprine from Babile district which is similar agro-ecological and similar animal rearing practice with our study, the result was 1.56% with RBT and 0.78 % with CFT of prevalence, this result also lower than in our study which is 11/181(6.07%) in C-ELIZA and 8/181(4.4%) with RT-PCR.

Ovine Brucellosis:

According to [29] review article on study of Brucellosis in Ethiopia in different species the prevalence of ovine with ELISA 9.7 and 9.4 results were identified in two Pastoral region of Ethiopia, this study was double of the two previous studies conducted which is about 20% of prevalence unlike the previous study.

Conclusion and recommendation

Our study identified *B. melitensis* and *B. abortus* were identified as causative agent of Camel, *B.melitensis* was for the causative agent of ovine and caprine brucellosis in the study area. Based on the study result screening of brucellosis should be practiced before an introduction of animal for breeding purpose and used as consumption of milk for human. In our study area camel milk is a primary food for the person who keep the herd, there is a direct contact of aborted fetus without any special care.

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CHAPTER 8. ABSTRACT FOR SCIENTIFIC PRESENTATION

8.1. Measles Outbreak in Mieso District, Oromia Regional State, Eastern Ethiopia, 10 October - 15, December, 2018.

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Key words: Measles, case control, Mieso.

Abstracts

Background: Measles is one of the commonly occurred disease in different part of Ethiopia with various morbidity and mortality rate, as Oromia health bureau received report from West Hararge zone of Mieso district we conduct outbreak investigation confirmation from December 20-25/2018, potential risk factor was identified and control measure was implemented.

Methods: A 1:3 unmatched case to control study were conducted. With 16 case and 48 control, a suspected case of any person with fever and maculopapular from Mieso district from October 10 to December 15, 2018. A line listed record was revised and a structured questioner was made from both case and control, all the record entered and analyzed with excel and epi.info 7.1.0.6 software.

Results: A total of 12 suspected and 4 conformed case were reported from October 10 to December 15, 2018, out of which 9 (56.25%) were female, with the mean age of 2.6 year (range from 6 month to 8 year), out of 48 control 34(70.8%) were female the mean age 40.8 years (range from 6 month to 48 years). Out of reported 4 kebeles Deneba hunde Misoma is the highest case reported 9(56.25%). The overall attach rate (AR) is 1.21% per 1000 population, zero case fatality rate.

Conclusion: Measles outbreak was identified, low immunization coverage and malnutrition were identified as a potential risk factor. All possible intervention and control measures were implemented.

8.2. Outbreak of Anthrax in Farta and Smada Districts of South Gondor Zone of Amhara Regional, 2019.

Abstract:

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Background;

Anthrax is a serious zoonotic disease caused by the bacterium Bacillus anthracis. We conducted an outbreak investigation to conform anthrax outbreak and describe cases.

Methods;

Descriptive case- control study design methods were used, samples were taken and an individual sample DNA extracted from Tissue with QIA amp® DNA extraction mini kit, amplification was done with Real time PCR for confirmatory test.

Result:

A total of 17 anthrax cases 5 (29.4%) female and 12 (70.5%) males with one death of female 45-year-old were reported from the two districts. An index case was a person with 55 years old reported on March 13, 2019. All samples taken from animal during March 13-April 8, 2019 were positive with Real time PCR. There was low vaccination coverage for livestock.

Conclusion

All cases were confirmed of Anthrax, the two districts were an endemic area of anthrax, there was a death before two years with similar situation, and there was low livestock vaccination coverage.

Recommendation

We recommend for the district health office to enhance the surveillance system so that anthrax cases could be identified earlier and increase livestock vaccination in both districts.

Annex 1: **Measles outbreak investigation form (Mieso District, West Hararge Zone, Oromia region**

Interviewee status: Case Control Date MM/DD/YYYY___/___/___/ ID No_____

A. Identification information:

1. Name_____ Region _____Zone_____

District _____Kebele_____

B. Socio-demography:

2. Age_____3. Sex: male/ female 4. Occupation_____

5. Respondent relation to cases or controls? Mother /Father / Guardian

6. Educational status of father: A. illiterate B.no Mother A, illiterate B. No

C. possible source of infection:

7. Have you travelled to somewhere else in the previous 7 to 18 days before the onset of rash? Yes No

8. If yes for No 8, where? _____

9. Contact with confirmed cases in the past 7 to 18 days? Yes No

10. If yes for No 10, where and who_____/_____respectively?

D. Clinical information:

11. Symptoms and signs: Fever Rash Cough Coryza Conjunctivitis

12. Date of onset of rash? MM / DD/ YYYY_____/_____/_____/

13. Have you visited health facility? Yes No

14. If yes for No 16, date seen at health facility____/_____/_____/

15. Complication Yes No

16. If yes for No 18: Diarrhea Ear infection Blindness Convulsion Other (specify)

E. Laboratory information:

19. Was sample taken? Yes No

20. If, yes, date taken____/____/_____/

21. Laboratory result: Positive for measles specific IgM Negative for IgM pending

F. Vaccination status:

22. Have you vaccinated for measles? Yes No

23. If yes, source of information? History Vaccination card

24. If yes for 25, last vaccination date____/_____/_____/

25. Dosage: One two & above don't know

26. No of children eligible for vaccination ---------& <yrs. -------- in the house hold?

G. Sanitation

27. Do you have latrine Yes/No

28. Do you have pipeline water supply yes/No

Thank you for your time!

Annex 2: **Questioner Survey for Anthrax Outbreak Investigation and Response**

Region _____ Zone_____

District_____ Kebele _____

Locality_____

Geo-reference:

Latitude_____Longtude_____Elivation_____masl

Name of Respondent_____ Age _____Sex_____

1. Farming System:
   Intensive _____ Semi-Intensive_____
   Extensive_____
2. How long you lived in this area_____
3. Do you know Anthrax _____if yes what is the local name for_____
4. Have you ever seen an Anthrax case either in Animal or Human? Yes _____No_____
5. If the answer is yes for question Number 4 when did you seen the case?
   _____
6. What are the main sign of Anthrax case?
   In Animal
   _____
   _____
   In Human
   _____
   _____
7. Do you think that Anthrax is treatable disease? Yes _____No_____
8. Did you vaccinated your animals against Anthrax Yes _____No_____
9. What measure had taken for anima with sudden death?
10. What measure to be taken post exposure with sudden animal death?

Thank you!!

Annex 3: Heath profile Data collection form Oromia Regional state Sebeta District

Name of district----------

Total area coverage ----------km square, urban---------- rural----------

Agro ecological zones

Geo- reference

Latitude----------longitude----------Altitude range --------- to ----------masl

Average annual rain fall ----------mm

Demography

Total population ----------male-------- female ----------

Urban inhabitant ----------male----------female----------

Rural inhabitant----------male----------female----------

Ethnic composition with in a district----------

6.2. Age composition

6.2.1 Children under 5 years

a. Male

b. Female

c. Total

6.2.2. 5-15 years

a. Male

b. Female

c. Total

6.2.3. 16-25 years

a. Male

b. Female

c. Total

6.2.4. 26-35 years

a. Male

6.2.8. 36-40 years

A. Male----------

B. Female----------

C. Total----------

6.2.9. 41-45 years

A. Male----------

B. Female----------

c. Total----------

6.2.9. 46-50 years

A. Male----------

B. Female----------

C. Total----------

6.2.20. 51-55 years

A. Male----------

b. Female

c. Total

6.2.5. 36-45years

a. Male

b. Female

c. Total

6.2.6. 46-55 years

a. Male

b. Female

c. Total

6.2.7. 56-65 years

a. Male

b. Female

c. Total

B. Female----------

C. Total----------

6.2.21 56-60 years

A. Male----------

B. Female----------

C. Total----------

6.2.22. 61-65 years

A. Male----------

B. Female----------

C. Total----------

6.2. 23. > 65 years

A. Male

B. Female 357(41.65%)

C. Total 857(.55%)

6.3. Number of pregnant woman----------

6.4. Number of lactating woman ----------

Economic status

No of Government employer

Male----------Female----------

Average monthly income ---------- range: highest------12338----- lowest------688---

Private employment----------

Average monthly income ----------range: highest----------Lowest----------

Daily laborer----------Male---------- Female----------

Average daily income ----------highest----------lowest----------

Education

No of formal education

KG/ primary education---------- Teacher student ratio----------

Secondary education----------Teacher student ratio----------

College----------Teacher student ratio----------

University----------Teacher student ratio----------

School drop out for the past five years.

8.5.1. Primary school

8.5.2 Secondary School.

Infrastructure

Transportation

Accessibility (Main road) yes----------no----------

Communication

9.2.1 No of people who had fixed telephone----------

9.2.1. No of people who access to mobile phone----------

9.3. Electricity

No. of house hold had power supply----------

Disaster status in area

No of disease out-break in district ----------

Type of disease----------

10.1.2. Laboratory confirmed ----------yes----------

Suspected----------

10.1.4.. No of cases ----------male----------female----------death----------male----------female

Age group

<5 years >45 years 23

15-40 years

Flooding ----------year of occurrence----------

No of people affected as a result of natural disaster ---------- male-------Female
Estimate direct effect on community property loss in Eth birr----------

Sanitation

11.1 sanitation coverage ----------%

11.2. Water supply coverage ----------%

11.3. Latrine coverage----------%

10.4. Latrine usage coverage ----------%

12. Vital statistics and human Indicator

12.1. No of still birth ---------- and in percent---------- (%)

12.2. Infant mortality rate----------

12.3. Child mortality rate----------

12.4. Crude birth rate----------

12.5 crude death rate ----------

12.6. Maternal mortality rate----------

12.7. Contraceptive prevalence rate ----------

12.8. Contraceptive acceptance rate----------

12.9. ANC Rate ----------

12.10. Percentage of deliveries attended by skilled birth attendants ---------- (%)

12.11. Percentage of deliveries attended by HEWs ---------- (%)

12.12. Average family size----------

13. Health service

Status of Primary Health Care Components

MCH

Immunization

Nutrition

Health Facilities.

Human resource distributions

Top ten cause of morbidity of Adults

Top Five mortality cause under five in an area?

Thank you!!

Annex 4: Public Health Surveillance evaluation Form

Region_____ Name of Respondent_____

Zone_____Tele_____

District_____Date_____

Availability of National surveillance Manual

1. Is there a national manual/ guideline for surveillance system?

Yes /No/ Not applicable / Unknown

2. *If yes*, describe (last update, diseases included, case definitions, surveillance and control, integrated or different for each disease):

_____

_____

3. What is the objectives of surveillance?

_____

4. What are the strengths of your surveillance system?

_____

5. What are the weakness of your surveillance system?

_____

_____

Ability to Case detection and recording

1. Do you have standard case definitions for the Country‘s priority diseases?

Yes / No / Unknown / Not applicable

2. If the answer is yes for Q #1, observe the presence of the standard case definition for each priority disease. Yes No

3. For which disease did, you lack the case definition?

_____

Reporting

1. What are the reporting entities for the surveillance system?

a. Public health facilities b. NGO health facilities c. military health facilities

d. Private health facilities e. Others_____

2. Means of communication in reporting from other source of health facilities

A. Fax B. Telephone C. Written hard copy D. Electronic copy E. by email

3. Did you have case based reporting formats for out breaks? Yes /No □ Not Applicable.

4. Was there guide line for specimen collection, handling and transportation to the next level? Yes/No □ Not Applicable

5. Did you have line list for reporting outbreaks? Yes/No □ Not Applicable

6. When are you expected to send weekly report to the regional PHEM unit?

□ Monday □ Tuesday □ Wednesday □ Thursday □ Friday □ Saturday □ Sunday□ I don‘t know

7. When are you expected to receive weekly report from HCs/HPs?

□ Monday □ Tuesday □ Wednesday □ Thursday □ Friday □ Saturday □ Sunday □I don‘t know

8. Did you have computer on your office? Yes/No

9. Did you have computer on your department (PHEM unit)? Yes /No

10. What is the data entry and compilation instrument? □ Manual □ Computer □ other_____

11. Did you have computer skill on □ MS word □MS excel □MS power point □ Epi-info

12. Did you analyze the data collected from surveillance system? Yes /No

13. How can reporting system be improved?

_____

_____

Outbreak investigation

Did you participate on outbreak investigation yes/ No

Do you have checklist for outbreak investigation? Yes/No

Did you report malaria as an outbreak? If yes When?

If you detect an outbreak where was laboratory confirmation of cases done?

□Regional laboratory □ Hospital □ EHNRI □ Health center □Contracted private laboratory □ Other_____

Do you think that your surveillance system could detect an outbreak? Yes/No/ No idea

If yes for question No.5 did you evaluate your system in such away? Yes/ No/ no idea

Do you have any comment for the improvement of your surveillance system
_____

Budget source

Did you received any budget for surveillance system? Yes/No

If yes for Q.1 from which source you received budget?
_____

If no. How do you overcome budget constraint??
specify_____

Thank you for your Time

Annex 5: Brucella project budget breakdown

Items	Activities	No person	No of days	Unit cost ETB	Total cost
Training	supervisor	1	2	40	80
	Data collector	1	12	30	360
Sampling	sample collection	90	4	50	18000
Hall rent	Hall rent	1	1	200	200
stationery	Note book, pen, flipchart, marker	1/once	per individuals	100	100
periderm for supervisor	supervisor	10	2	40	800
data collector	Data collector	15	12	30	5400
laboratory consumables	reagents, primers, probes, Elisa kits, and consumables	reagents and consumables	sum /items		20,000
Bench fee	for Lab personnel	30	4	30	3600
Allowance for focal personnel	facilitate, coordinate, data collection and writing results	1	12 month	300/month	3600
Transport	car rent	90 days	1	100	9000
Total					61140

Annex 6: Epidemiological and Molecular study of brucellosis form

Dear respondent,

This questionnaire intends to obtain data on *risk factors associated with Brucellosis in* your area. Your cooperation to respond for this questionnaire is very important to this research survey because it represents hundreds of others who were not included in the sample. Please respond to every question accordingly. Thank you in advance for your cooperation

Date_____ Name of the farm/ farm owners name _____

Phone no. _____

Farm ID: _____

For individual animal ID (Team code and number example team 01, = 01-0001,

Address: Region _____ Zone_____ Woreda _____Kebele_____

Specific place/Village name_____

Geographic reference of the farm/village:

Longitude _____ Latitude _____ Altitude _____

What species of animal do you keep?☑

Bovine Ovine ☐ Equine ☐Caprine ☐Camel ☐others ☐

Herd size of the farm?

☐Small (up to 10) ☐ Medium (11-20) ☐Large (>20)

What is the livestock production system? ☑

Intensive management system

Semi Extensive management system (Mixed crop livestock)

Extensive management system (Pastoralism)

4. How do you keep your livestock? ☑

Separating all species

Keeping all together

5. How often do you clean the house of your livestock? ☑

☐ Daily ☐ Twice a day ☐every week Monthly ☐ Others

6. Did you use artificial insemination? ☑

Yes

No

7. If the answer of Q6 is No what type of breeding system you use? ☑

Bulling system

Natural breeding

Others

8. Do you have calving pen / paddock?

☐ Yes ☐ No

9. Is there a history of abortion or stillbirth?

☐ Yes ☐ No

10. How many animals aborted within one year duration (Since last year, by species cattle, sheep, goat, camel?)

Species	abortions/still births in the last 1 year
If camels present	
If cattle present	
If sheep present	
If goats present	

11. If the answer for Q No 9 is yes, in which trimester?

1st trimester

2nd trimester

3rd trimester

12. Where do you dispose the aborted fetus/retained placenta during normal delivery? ☑

Incinerating/Burning

Disposing to the environment

Deep burring

Allow to be eaten by carnivores

13. If your animals faced a problem during parturition who is going to assist animal? ☑

Veterinarian/Para veterinarian/CAHWs

By your self

Traditional healers

Others

14. If you assist parturition/delivery yourself, do you use glove:

Use glove,

Bare hand without hand protection

15. What do you usually do with the animal that have frequent abortion?

Sell to other farms, sell to slaughter, ☐ and give as gift, ☐

Others

16. Have you introduced new cattle (purchase, gifts etc) ☑

Yes

No

17. If yes, any test for diseases like brucellosis before introduction to farm?

Yes No: if yes

18. What do you did with positive animals:

Sell for other farms, ☐

Home slaughter, ☐

Sell for slaughter, ☐

Remain in the farm with no measure,

Others please specify)

19. Are there wild animals in the area? ☑

Yes

No

20. Is there close contact between wild life and livestock while grazing or watering? ☑

Yes

No

If yes, have you ever found evidence of wildlife abortion in your area/ where you graze your animals?

 Yes

 No

21. Is there any disease outbreak which cause abortion, stillbirth, arthritis, orchitic etc. in your area currently? N.B especially abortion in late pregnancy ☑

Yes

No

22. Do you know the local name of the disease? ☑

Yes

No

If "yes "_____

23. What is the main way meat is typically consumed in the household??

Raw

Cooked/boiled

Both

24. What is the main way milk is typically consumed in the household? (Please select one of the following, separate ways for children and adult)

 Raw milk

 Boiled milk

Thank you!!