EPIDEMIOLOGICAL INVESTIGATION OF CUTANEOUS LEISHMANIASIS IN SODO DISTRICT SOUTHERN ETHIOPIA

by

LINA GAZU MEGO

Submitted in accordance with the requirements

for the degree of

DOCTOR OF LITERATURE AND PHILOSOPHY

in the subject

PUBLIC HEALTH

at the

UNIVERSITY OF SOUTH AFRICA

SUPERVISOR: PROF ZERISH ZETHU NKOSI

CO-SUPERVISOR: DR NIGATU KEBEDE

October 2021

DEDICATION

To my Yahweh Rohi (Lord my Shepherd) and Yahweh Yireh (Lord my provider) Jesus Christ: who always push me forward against all odds.

DECLARATION

Name: Lina Gazu Mego

Student number: 57647542

Degree: Doctor of Literature and Philosophy

EPIDEMIOLOGICAL INVESTIGATION OF CUTANEOUS LEISHMANIASIS IN SODO DISTRICT SOUTHERN ETHIOPIA

I declare that the above thesis is my own work and that all the sources that I have used or quoted have been indicated and acknowledged by means of complete references.

I further declare that I submitted the thesis to originality checking software and that it falls within the accepted requirements for originality.

I further declare that I have not previously submitted this work, or part of it, for examination at Unisa for another qualification or at any other higher education institution.

October 31, 2021

Lina Gazu Mego **Date**

ACKNOWLEDGEMENTS

First and above all, I honor God, the almighty for giving me this chance and granting me the ability to progress well.

This thesis would not have been possible without the support and assistance of a number of persons who contributed in one way or another to the preparation and completion of this research.

My primary as well as especial appreciation goes to my supervisor, Professor Zerish Zethu Nkosi, whose genuineness and inspiration I will never fail to remember for she has been my muse as I hurdle all the obstacles in the completion of this research project. I am also heartily thankful to my co-supervisors, Dr Nigatu Kebede, for availing his unreserved support in several ways.

I'd like to express my heartfelt gratitude to Sebseba Kebede for assisting with field operations, as well as the data collectors who actively participated in the study, shared their experiences, and contributed to the study's success. I also express my thankfulness to the study members who take part in all phases of the study. I am incredibly grateful to Armauer Hansen Research Institute (AHRI) and experts working in Leishmania laboratory – Dr Endalamaw Gadisa and Demekech Damte for their invaluable help during the lab work. I also offer my regards to UNISA and Addis Ababa University, for the financial support.

This thesis would not have been possible without the help of my dear husband Elias Abate and my lovely son Yohanan Elias. I appreciate greatly and dearly the endurance and patience paid by them and I owe my deepest gratitude to their morale support throughout the study period.

Lastly, I do not want to windup my ackneowledgement with out offering my regards to all individulas who offer their invaluable support for the completion of the project.

EPIDEMIOLOGICAL INVESTIGATION OF CUTANEOUS LEISHMANIASIS IN SODO DISTRICT SOUTHERN ETHIOPIA

STUDENT NUMBER: 57647542

STUDENT NAME: Lina Gazu Mego

DEGREE: Doctor of Literature and Philosophy

DEPARTMENT: Department of Health Studies

SUPERVISOR: Prof Zerish Zethu Nkosi

CO-SUPERVISOR: Dr Nigatu Kebede

ABSTRACT

In Ethiopia, Cutaneous Leishmaniasis (CL) is a common infectious disease. However, existing knowledge on prevalence, risk factors, community awareness and circulating parasite species are scarce. The study aimed at producing evidence to inform prevention strategies through epidemiological study using different approaches. Objectives are to access knowledge, attitude, and practices about CL, investigate risks related to transmission and determine prevalence of CL in Sodo District. Primary quantitative method using a cross-sectional descriptive approach was applied. Data were collected from interviews held with 423 households for the KAP survey. In factor assessment and prevalence study, 379 households harbouring 1356 individuals were interviewed along with clinical assessment.

In the KAP study, 263 (61.9%), 226 (53.4%), and 213 (50.4%) have satisfactory knowledge, favourable attitude, and good practice about leishmaniasis. Majority are unable to identify leishmaniasis and unaware of its cause and transmission, respectively. Sandfly is considered "important biting and blood-sucking insect" by 210 (49.6%) but knowledge on biting time and breeding place was unsatisfactory. Most believed CL can be treated, is a serious problem and have a disfiguring outcome. However, most (59.3%, n=251) prefer use of traditional medication. Concerning practice, 288 (68.1%) have bed nets but personal protective measures are rarely used.

Risk factor assessment shows kebele, presence of screen on window/door, sensel tree and cave/gorge, host factors (travel history, sleeping at home yard, irrigating at night, spending time near cave/gorge, using personal protective measures and sleeping hand/face covered), hyrax near home and dumping dung to be significant factors affecting leishmaniasis.

Prevalence of leishmaniasis was 9.07% (123/1,356) for both scars (5.45%) and active (3.61%) infection. Lesions were mainly located on face parts and LCL was the most common form. The PCR test identified *Leishmania aethiopica* as principal species.

The Sodo community will benefit from awareness creation programs cause, transmission, and vector of leishmaniasis. Prevention programmes should prioritise changes in host behavior such as putting a screen on window or door, use of personal protective measures, irrigation at night, and sleeping covering hand and face. CL is a challenging public health crisis in the district and due attention needs to be given to it.

KEY TERMS: Attitude; Cutaneous Leishmaniasis; Ethiopia; knowledge; Leishmania species; Lesion; Practice; Prevalence; Risk factors; Sodo District; Transmission; Vector-borne disease.

Table of content

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

AIDS:	Acquired Immunodeficiency Syndrome
ALERT:	All Africa Leprosy Rehabilitation and Training Center
ALIPB:	Aklilu Lemma Institute of Pathobiology
AVL:	Anthroponotic Visceral Leishmaniasis
BCC:	Behaviour Change Communication
CDC:	Disease Control and Prevention
CL:	Cutaneous Leishmaniasis
CSA:	Central Statistical Agency
DAT:	Direct Agglutination Test
DCL:	Diffuse cutaneous Leishmaniasis
DDT:	Dichloro Diphenyl Trichloroethane
DNA:	Deoxyribonucleic acid
ELISA:	Enzyme-Linked Immunosorbent Assay
HIV:	Human Immunodeficiency Virus
IFAT:	Indirect Fluorescent Antibody Test
IRS:	Indoor Residual Spraying
KAP:	Knowledge attitude and practice
LCL:	Localized Cutaneous Leishmaniasis
LST:	Leishmanin Skin Test
MCL:	Muco CutaneousLeishmaniasis,
MCL:	Mucocutaneous Leishmaniasis
NNN:	Novy- Nicolle –McNeal
NTDs:	Neglected Tropical Diseases
PCR:	Polymerase Chain Reaction
PKDL:	Post kala-azar dermal leishmaniasis
SNNP:	Southern Nations, Nationalities, and Peoples'
SPSS:	Statistical Package for Social Science
VL:	Visceral Leishmaniasis
WHO:	World Health Organization
ZVL:	Zoonotic Visceral Leishmaniasis

LIST OF ANNEXURES

CHAPTER 1

ORIENTATION OF THE STUDY

1.1. INTRODUCTION

Leishmaniasis is a parasitic disease caused by flagellated protozoans of the Leishmania genus. It is spread by the bite of a female hematophagous sandfly vector, which is widespread in humans and certain animals (anthropozoonoses). Leishmaniasis is prevalent in nearly a hundred countries, resulting in millions of new cases and up to 65,000 deaths per year (Omari, Chahlaoui, Talbi, Ouarrak & Lalami 2020:1). Cutaneous Leishmaniasis (CL), mucosal involvement (MCL), and systemic visceral involvement (VL) are all clinical manifestations of the illness in humans (Eshetu & Mamo 2020:1).

CL is the most common type of leishmaniasis worldwide. CL affects over one million people worldwide per year, with hotspots in Afghanistan, Algeria, Iran, Pakistan, Peru, Brazil, Saudi Arabia, Colombia, and Tunisia (Akram, Khan, Qadir & Sabir 2015:1). Due to environmental changes, host immune status treatment failure and drug resistance, re-emergency of CL is recently observed in many endemic countres (Bekele, Bekele, Mulatu, et al. 2014:2).

In Ethiopia, the yearly burden of CL is eatimated to be between 20,000 to 30,000 cases putting the country among high CL burden countries (WHO 2014). *Leishmania aethiopica* is the most important cause of CL in Ethiopia (Henten, Adriaensen, Fikre, et al. 2018:69). The interesting features of leishmanaisis caisued by *L. aethiopica* are atypical lesions appearing commonly on the face, healing of lesion taking longer time and lesions are more severe in their presentation than that of other species (Henten et al. 2018:69).

The first CL case in Ethiopia was reported in the early 20th century. "Kunchir" in Gojam, "Finchoftu" in northern Shoa, Gonder, and sections of Wollo, "Chewie" in Sodo, "Shahegne" in north Shewa, "Volbo" in Ocholo, "Giziwa" in Tigray, and "Simbirahalkani" in Wollega are some of the local names for CL (Tilahun, Alemu & Mulatu 2014:190). CL is endemic in Ethiopian highlands with an altitude > 2,000

meters (Dassoni, Daba, Naafs & Morrone 2017:106). CL has been estimated to threaten about 30 million Ethiopians, predominantly from the highlands of Amhara, Tigray, Oromia, and the Southern Nations, Nationalities, and Peoples' Region (Seid et al. 2014:377).

Disfigurement from CL cause both social and psychological influences such as nervousness, tension and depression leading to low quality of life which in trun affects economic productivity of individuals (Tamiru, Mashalla, Mohammed & Tshweneagae 2019:1). However, adding to the growing proof of an evolving challenge, no vaccines exist for human, and difficlulty in treatment, particularly in resource-limited countries like Ethiopia (Seid et al. 2014:378). Data on precise level of CL is deficient in Ethiopia and the country has not desined a program to control the vectors transmitting the disease (Yohannes, Abebe & Boelee 2019:2). Moreover, the disease is the most overlooked disease in Ethiopia among all tropical diseases neglected so far. There is an urgent need of epidemiological investigation of in the country due to lack or inadequate diagnosis and therapy access and limited proof, or management attempts of CL in the country (Yohannes, Abebe & Boelee 2019:2). As a result, controlling CL necessitates an integrated approach that takes into account risk factors at both the individual and environmental levels. The Ethiopian Federal Ministry of Health has established a foundation to construct a leishmaniasis control program, recognizing the growing burden of the disease. In order to control and prevent leishmaniasis in Ethiopia, the Ministry has also designated vector control, health education, social mobilisation, and information system strengthening as priorities (Guideline for Diagnosis, Treatment & Prevention of Leishmaniasis in Ethiopia, 2013).

1.2. BACKGROUND TO THE RESEARCH PROBLEM

Understanding of CL by local population is one of the key elements that determine achievement of management programme. Community awarness is vital to know symptoms for effective case management. Infection could be prevented by abiding with protective methods like reducing contact with reservoir animals and avoiding sand fly bite by wearing protective clothes to cover exposed body parts during

outdoor works. Additionally, using insecticide-impregnated nets and managing household settings to reduce sandfly size are preventative measures (Wijerathna, Gunathilaka, Gunawardana & Rodrigo 2020:5). Awareness creation on how CL spread including reservoirs and vectors, the focal time when sandfly bite, and how to avoid the bite and decrease the sandfly populations should also be part of the prevention plan (Wijerathna, Gunathilaka, Gunawardana & Rodrigo 2020:5). Assessments have demonstrated that the existence of sufficient understanding has a substantial impact on success of control programmes via modifications in behaviour and more extensive involvement (Costa, Marques, Ramos, Santana, Alves & Faustino 2013). Before planning any awareness creation programme, it is vital to gather evidence on the level of existing knowledge and gaps in the community. This evidence will serve as an indicator for health education and promotion activities, identify research gaps, and feed data for planning applicable policies to monitor government and community attempts. Nevertheless, there are few KAP surveys in the country and none in the study area to the knowledge of the researcher.

Each state and region in Ethiopia have a different climate, topography, weather, living, and even house construction styles. This will affect the transmission dynamics of diseases particularly those greatly dependent on environmental and behavioural factors. Varied factors such as vector, reservoir, host, and the environment affect the biology and transmission cycle of leishmaniasis (Oryan & Akbari 2016:925). Environmental factors such as temperature fluctuations, irrigation habits, water storage, desertification, immunosuppression by HIV or organ transplant, climate changes, drug resistance, travel to CL-endemic areas, and socio-cultural factors such as war and low socio-economic status all influence the spread of leishmaniasis (Oryan & Akbari 2016:925). Additionally, it was shown that the spread progression of Leishmania displays attributes that are unique to every CL widespread region (Tamiru et al. 2019:9). Therefore, it is vital to understand factors playing a part in spread dynamics of the disease in a particular area. So far, factors cpntributing to the transmission of CL in Sodo district are not investigated.

Patient examination and treatment of leishmaniasis are dependant on the clinical and compatible epidemiologic context. So, verification and recognition of the species of *Leishmania* are important (Torres-Guerrero, Quintanilla-Cedillo, Ruiz-Esmenjaud & Arenas 2019:10). Clinical presentation of infection triggered by various species of leishmaniasis has a particular future (Griensven, Gadisa, Aseffa, et al. 2016:3) which has implication for treatment approaches. Since species-specific treatment is needed and to design a feasible prevention/control strategy, a study to identify species revolving in the community is mandatory. Leishmania species circulating in the community is not studied in Sodo district.

Sodo is a woreda in Ethiopia's Southern Nations, Nationalities, and Peoples (SNNP) Region. The region is one of the four areas where CL is known to be endemic (Seid et al. 2014:377). In 2005, outbreak of CL was reported from Silti District (Negera, Gadisa, Yamuaha, et al. 2008) which is 158.6 km from Sodo. It is obvious that Sodo is at risk of CL and there is a need for research to find the size of the illness and pinpoint circulating Leishmania species in the district.

1.3. RESEARCH PROBLEM

Even though CL is a widespread condition in Ethiopia, little is known about its impact. According to studies, the disease affects around 0.5% of the total population in the highlands. So far, no epidemiological data have been reported concerning the prevalence of CL in Sodo although a prevalence of 4.82% was reported in Silti District which is close to Sodo (Negera et al. 2008). It has been almost ten years since the study in Silti was conducted. This study, thus, will be essential in highlighting the spreading of CL and delivers the knowledge gaps regarding CL in the district. Furthermore, this research will give information on the extent of CL in the Sodo District. Prevalence study of CL will help design of proper control methods and understand risk status of the community. Consequently, this research evaluated the level of CL and the role of varied factors in Sodo districts of SNNP state. Ethiopia is one of the countries where there is a limited resource for the prevention of diseases. Evidence-based disease control and prevention strategies need to be designed for allocation of budgets. As a result, this study provides information about

the disease's control priorities in the area. A fundamental component of a disease control program is community education on illness prevention and control. It is vital to understand knowledge gaps in order to put such awareness-building initiatives into action. As a result, the current study aimed to assess the Sodo community's CL knowledge, attitudes, and practices.

There is neighter vaccination nor an effective treatment scheme for the treatment of CL. The few drugs available take two weeks for treatment and may even extend to three weeks or more if there is mucoCL. Additionally, these drugs are unavailable to the rural communities as the centres offering diagnosis and treatment are few in Ethiopia. Therefore, prevention is the main strategy to tackle the disease. To design effective prevention programmes, specific factors affecting the transmission of the disease must be defined. This is vital because factors for CL transmission vary from place to place. In cognizant of these, this research intended to navigate through different factors contributing to the disease transmission.

To take proper actions against CL, the magnitude of the disease should be estimated. In addition, for proper treatment of CL, the Leishmania species exciting in the area should be identified because the treatment of CL is species-specific. Putting these into consideration, this study focused on investigating the prevalence of CL and identifying Leishmania species in the study area.

1.4. PURPOSE AND AIM OF THE STUDY

The goal of this study was to add to the existing body of knowledge and enhance the lives of the Sodo community by gathering epidemiological data and creating evidence for designing effective leishmaniasis control and prevention strategies.

The aim of the study is investigate CL transmission in Sodo District using different approaches (Knowledge attitude and practice survey, risk factor assessment, and finally prevalence estimation and parasite identification).

1.5. RESEARCH OBJECTIVES

The following specific objectives were studied in order to achieve the desired goal::

Phase 1: -

- To access knowledge, attitude and practices of Sodo community on CL.

Phase 2: -

- Investigate demographic, household and host-related risks related to transmission of CL.
- To clarify the role of animal and environmental factors on spread of CL in the district.

Phase 3: -

- To determine the prevalence of CL in the area.
- To identify the Leishmania species circulating in the community.

And generally, to understand key features leishmaniasis transmission in the community and highlight ways forward for the managing CL in Sodo District based on results-driven from objectives above.

1.6. RESEARCH QUESTIONS

- What is the knowledge attitude and practice of the society about CL?
- Which demographic, household and host-related risks related to transmission of CL in the area?
- Which animal and environmental factors affect spread of CL in the district?
- What is the prevalence of the disease in Sodo District?
- What species of Leishmania is circulating in the community?

1.7. SIGNIFICANCE OF THE STUDY

- CL is a very important health problem worldwide and particularly in Ethiopia and infection may occasionally the lesion may result in disfiguring scars with a lifelong aesthetic stigma.
- The results of this study will present evidence on
    - Prevalence of CL in the Districts.
    - Knowledge, attitude, and practices of the society about CL.
    - Factors adding to the transmission of the disease.
    - Identification of Leishmania species circulating in the community.

  These data will help develop a feasible strategy to control CL.

1.8. DEFINITION OF KEY TERMS

- Cutaneous Leishmaniasis - "is an endemic illness spread by the sandfly and characterised by the formation of cutaneous papules that develop in nodules, break down to produce ulcers, and heal on their own, leaving a scar." (WHO, 2020). CL is an unpleasant and stigmatising disease that can range from a modest self-healing cutaneous lesion to significant cutaneous and mucocutaneous involvement (MucoCL, MCL, and CLDCL, DCL) (WHO. 2020).

- CL case definition-

  A suspected case can be defined as an individual demonstrating clinical symptoms (skin lesions) like papule which develop to ulcerated nodule or plaque which then persists for a varying time prior to self-healing leaving a scar (Glans et al. 2018).

  A confirmed case is when a suspected case is coupled with parasitological proof of the examination (on smear or culture) (WHO 2020).

- Vectors - "living organisms capable of transmitting infectious illnesses between humans or from animals to humans." (WHO, 2020). Several of them are vampiric flies that absorb pathogenic germs while feeding on the blood of an infected host and then transfer the pathogen to a new victim once it has

reproduced. In most cases, vectors propagate the virus for the rest of their lives during each bite/blood meal (WHO, 2020).

- Vector-borne disease – "Vector-borne diseases are human illnesses caused by viruses, parasites, or bacteria transmitted by vectors." Dengue fever, malaria, schistosomiasis, leishmaniasis, human African trypanosomiasis, Japanese encephalitis, Chagas disease, onchocerciasis, and yellow fever kill around 700,000 people each year." (WHO, 2020). The burden of vector-borne diseases is largest in the poorest populations in tropical and subtropical areas. A complex collection of demographic, socioeconomic, and environmental factors, including worldwide travel and trade, as well as spontaneous expansion, determine the spread of vector-borne diseases (WHO 2020).

- Prevalence study – Or cross-sectional study is the most common population-based epidemiological studies created to measure the incidence of a health event in the people at a moment in time or over a short period of time are prevalence

  A prevalence rate (P) is valued by:

  P = Sum of individuals with a disease

  The inhabitants at risk of developing the disease in a given time

  The population at risk can be a population living in the study area or can be specified by geographical, administrative, demographical, occupational, or other parameters, such as health services clients. The prevalence rate is reported on a population base, e.g., 5 cases of a disease per 100 peoples (5%) (Spronk, Korevaar, Davids, Hilderink, Schellevis, Verheij & Nielen 2019).

- Knowledge attitude and practice (KAP) study – A Knowledge, Attitude, and Practices (KAP) survey is a quantitative method that helps gather quantitative and qualitative information using standardised questionnaires.

KAP surveys are used to identify and describe obstacles to behaviour changes by pinpointing fallacies (Monde 2011). KAP surveys are important to examine discrepancies between what is said and what is done (Monde 2011).

- Risk Factor – is any element, trait, or contact of a person that rises the probability of acquiring a disease or injury (Ghezzi 2020:2). Risk factors for CL could be the presence of hyraxes or vectors near residents, working outside at night, the presence of bed nets in the house, sanitation, and hygiene.

1.9. OPERATIONAL DEFINITIONS

- Skin test:- is “inspection of slit skin smears for amastigotes by taking tissue material from the inflamed and swollen border of an ulcer or nodule, but not from its base or center, which typically holds only dead tissue with care to avoid contaminating the specimen with blood” (Desta, Shiferaw, Kassa, Shimelis & Simachew Dires 2005).

- Culturing on Novy- Nicolle –McNeal (NNN) medium: - is a familiar and widely used medium for identification of species in genus *Leishmania* which gives rise to the motile and extracellular promastigote form of the parasite. Novy, McNeal developed the medium and later modified by Nicolle. This medium comprises of blood agar which encourages the development of meticulous organisms like *Leishmania* and *Trypanosoma* and an overlay medium (called Locke’s solution). Following incubation for 24 hours in the liquid part of the medium, the amastigotes convert to promastigotes (Tankeshwar 2016).

- Parasitological examination: - is a laboratory method in which Leishmania Parasite can be displayed after staining (Giemsa’s staining method Or Field’s staining technique) and /or culture technique (Desta et al. 2005).

- Clinical examination: - This is a test that looks for symptoms of illness in people who are suspected of being infected. CL stands for localised cutaneous lesions that have no link to the mucosa and are not a generalised sickness. The hands, face, forearms, and lower limbs are among the vulnerable regions of the body that sandflies can access. Lesions may form and become disseminated CL in rare cases, with many nodules that begin as erythematous papules and grow to their final size in a few weeks. A well-defined, numerous, round to oval regular contour with varied dimensions (0.5–10 cm on diameter) distinguishes disseminated CL (Desta et al. 2005).

- Knowledge: - Public comprehension of CL is referred to as knowledge. The mean score of the eight knowledge elements is used to quantify it. Knowledge is classified as satisfactory (if participants earned a mean score of the properly answered questions) or unsatisfactory (if participants scored a mean score of the correctly answered questions) in this study.

- Attitude: - This refers to how the general population feels about CL and how they act in response to it. Six attitude questions are used to calculate it. Overall scores were generated from all personal responses to attitude questions, and the mean score was used to categorise participants as having a favorable attitude (if participants scored a mean score) or an unfavorable attitude (if respondents scored a mean score).

- Practice: - is participation in the community on a regular basis to avoid CL. It is graded on the basis of eight practice questions. All individual responses to practice questions were analysed to get overall mean scores, which were then classified as excellent practice (if respondents scored above the mean) or poor practice (if people scored below the mean).

1.10. THEORETICAL FOUNDATIONS OF THE STUDY

Environmental, Behavioural, immunogenic, genetic and socio-economic factors put human populations at risk (Reveiz, Maia-Elkhoury, Nicholls, Romero, & Yadon 2013:2). Houses with fractured mud or thatched covered walls, wet mud floors, and vegetation close to the house can accelerate sandfly existence and improve vector profusion by offering daytime resting spots, reproducing sites and moistureLack of pesticide application in homes has been linked to a higher risk of illness (Coura-Vital, Reis, Reis, Braga, Roatt, Aguiar-Soares, Marques, Veloso & Carneiro 2011:411). Sleeping on the floor or outside can expose to leishmaniasis but insecticide treated bed nets is very crucial for people in safeguarding against leishmaniasis transmission (Voty'pka, Kasap, Volf, Kodym & Alten 2012:188). Figure 1.1 shows the theoretical context of the interaction of factors affecting CL.

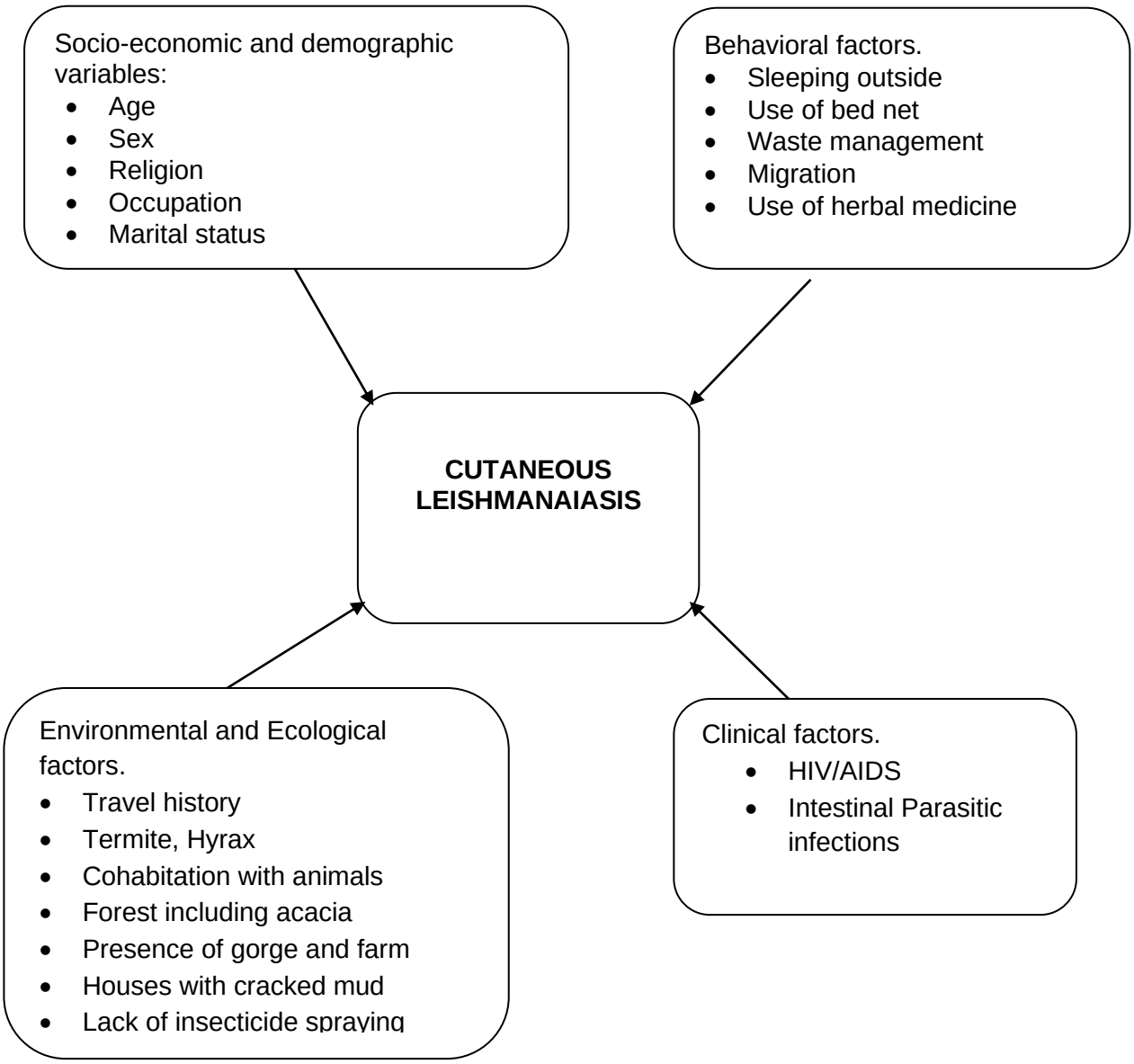


Figure 1.1: Theoretical context of exposing factors for CL transmission (Adopted from Dawit et al. 2013 & Negera et al. 2008:886).

Migration of people from rural to urban parts owing to get better quality life and attain better social services or socio-economic circumstances and unsuitable climate or even relocation to communities can upsurge cases of CL (Reithinger, Mohsen, & Leslie, 2010:6). Environmental factors such as elevation, new agricultural projects, closeness to woodland, forest coverage, irrigation, waste

products storage near city can increase in sandflies population (Valderrama-Ardila, Alexander, Ferro, Cadena, Marín, Holford, Munstermann, & Ocampo 2010:246).

Rodents and dogs existence is a major risk factor for CL as these play part in the spread cycle of Leishmania (Belo, Werneck, Barbosa, Simo˜es, Nascimento, Silva & Struchiner 2013:3). The presence of animals especially cows could increase leishmaniasis risk as cow manure gives a fertile ecosystem for the sand flies attracting them nearer to people (Bern, Courtenay & Alvar 2010:2).

Intestinal parasitic diseases triggering malnourishment and HIV are added risk factors sensitising individuals to leishmaniasis (Mengesha, Endris, Takele, Mekonnen, Tadesse, Feleke, & Diro 2014:5). Undernourishment, minimal nutritional protein, vitamin A, iron, zinc, and energy levels boost the risk of VL and mucoCL. This effect is due to the lymph node's malfunctioning operation and the Leishmania's premature visceralisation (Dawit, Girma & Simenew, 2013:3). Different susceptibility to CL may be caused by gender differences. Because of the role of sex hormones in modulating the reactivity to Leishmania, women are at a lesser risk (Mengesha et al 2014:5). Furthermore, males are increasingly spending time outside the home performing agricultural tasks, and sand flies may bite them in rural locations (Mengesha et al. 2014:5).

1.11. RESEARCH METHODOLOGY

1.11.1. Research approach

Quantitative method was employed in this study. Quantitative approaches are used to measure objectives through numerical, statistical, or mathematical assessment of data from questionnaires, and surveys (Babbie 2010:90-107). Numerical data collected through quantitative research will help to have a boad view of groups of people or understand difference in disease status among individuals. Quantitative research study is done with aim of classifying and counting characteristics then building a model to explain observations (Rich, Brians, Manheim & Willnat 2011:75-87).

Quantitative research can be calssified in to correlational, descriptive, experimental and causal-comparative/quasi-experimental research. Descriptive quantitative study examines variable of interest. Hence, through descriptive research systematic information about phenomenon obtained. However, in correlational research, the scope of a connection among variables is explained applying statistical data (Winston-Salem State University, 2020). The existence of a correlation or relationship between variables is not always indicative of causation. Causal comparative/quasi-experimental research is used to identify cause-effect correlations between variables. An independent variable is observed in comparative/quasi-experimental research, but it is not controlled by the investigator as it is in experimental investigations. Rather, the impact of the independent variable on the dependent variable is evaluated. The study is experimental research if scientific methods such as treatment administration are used to determine the cause-effect relationship between a collection of factors (Winston-Salem State University, 2020).

1.11.2. Research design

In this study, a primary quantitative method with a cross-sectional descriptive approach is used to assess the community's knowledge, attitude, and practice regarding leishmaniasis, as well as to assess different risk factors that contribute to the spread of CL and to investigate the disease's prevalence in the area. Data is collected at a specific point in time in a cross-sectional study (Von Davier& Lee 2019).

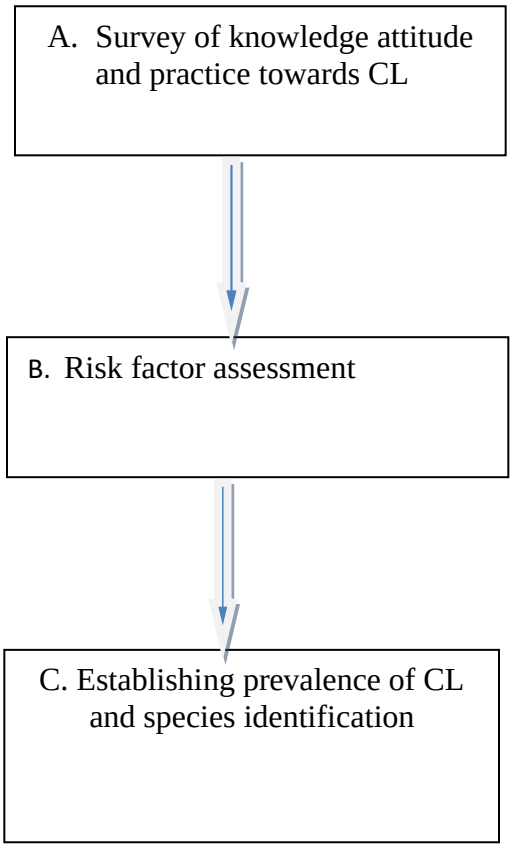


Figure 1.2: Flow of research phases.

1.11.2.1. Analysis of knowledge attitude and practice towards CL

A population-based cross-sectional survey was conducted in the Sodo District of Ethiopia's SNNP region to investigate CL-related KAP among the Sodo people. A questionnaire with structured questions was constructed in order to provide information on socio-demographic characteristics, CL knowledge, CL attitude, and CL practice in the English language.

1.11.2.2. Risk factors assessment

A structured questionnaire will be created to evaluate the risk variables for CL transmission. Household variables, environmental and ecological factors, host factors, and animal factors will be divided into four categories.

1.11.2.3. Establishing the prevalence of CL and species identification

A house-to-house assessment was conducted to get cases of CL in the selected Kebeles in the district. A separate questionnaire was administered, and a patient datasheet was formed. Biopsy and skin scrapping samples from clinically suspected subjects for CL by a senior nurse. Exudates were smeared on a plain glass slide, fixed with methanol, and stained with Giemsa for parasitological analysis (Geimsa stain). Finally, the slides were microscopically examined and compared to a positive control slide. Following adequate sample treatment, a variety of laboratory tasks were carried out, including:

- Parasite Culture on Novy-MCNeal-Nicolle (NNN) media
- Species identification (Polymerase Chain Reaction)

1.11.3. Study setting and population

The research was carried out in the Sodo District. Sodo District is situated in Garage Zone of SNNP's Regional State. The geographical location of the district according to Ethiopian Central statistical agency, is from 8° 09' to 8° 45' North latitude and from 38° 37' to 38° 71' East longitude (CSA, 2007). It is located approximately 103 kilometers south of Addis Ababa on the route. The Woreda has an area of 88,553.3 hectares. The Woreda also shared its boundary with Meskan Woreda on the south, on the west, north, and the east by Oromia Region.

The population for this study is community dwellers in the district.

1.11.4. Sample and sampling methods

Based on traditional healers' consultation, District health professional's direction, and secondary data analysis, six kebeles were selected as a study area based on the burden of CL.

A. Survey of knowledge, attitude, and practice towards CL: Sample size for the questionnaire study was computed assuming 50.0% knowledge of community on CL with a marginal error of 2% and 95% confidence level.

Adding up a 10% non-response level gives a final number of households to be visited for this survey was 423.

B. Risk factors assessment and prevalence study: since there was a gap of six months between the KAP survey and lab data collection and many other factors we faced (Internal unrest), we were forced to select another population of participants for risk factor assessment and prevalence study using the same calculation in the KAP survey. Therefore, another 423 households were visited for this survey.

1.11.5. Data analysis

- For KAP survey frequency, percentages, chi-square test, and univariate and multiple logistic regression were performed using SPSS version 24.
- For risk factor assessment frequency, percentages, univariate and multiple logistic regression, multiple correspondence analysis, and hierarchical cluster analysis were done using R statistical package 3.0.
- For prevalence study frequency and percentages were applied.

1.11.6. Validity and reliability

To achieve validity and reliability, several efforts were made such as expert consultation, pretesting of data collection tools, use of standardised questionnaire, defining variables, random selection of participants and use of proven methods by other studies.

1.12. ETHICAL CONSIDERATIONS

The research was approved by the Ethics Committee of the University of South Africa's Department of Health Studies. The SNNPR health bureau received a letter from Unisa Akaki Regional Learning Centre demonstrating the research's goal. The SNNPR health bureau, Gurage zone administration, and Sodo Health office all issued approval letters.

Adherence to standards of autonomy, beneficence, non-maleficence and justice was assured to protect both the researcher and participants during the investigation.

1.13. SCOPE AND LIMITATION OF THE STUDY

- The scope of the research goes from gaining access to community indigenous knowledge to investigating the disease's transmission dynamics (factors and parasite identification) in the Sodo District in South Ethiopia.
- Research limitations: the study area is limited to the Sodo district and does not reflect the entire region (SNNP region).

1.14. STRUCTURE OF THE THESIS

The structure of the thesis was partitioned into the following chapters:

- Chapter 1: Orientation to the study – gives overview of the study and structure of the thesis.
- Chapter 2: A literature review of the study – discusses current knowledge about CL and trends in the world.
- Chapter 3: Research design and method – illustrates methods used for gathering data and strategies applied for achievement of study objectives.
- Chapter 4: Analysis and Presentation of results – Results form the three phases are explained in detail.
- Chapter 5: Discussion of the result – findings are discussed against other similar studies in the world and in Ethiopia.
- Chapter 6: Conclusions and recommendations – this chapter highlight ways forward for the managing CL in Sodo District based on results-driven from rstudy findings.

1.15. SUMMARY OF STUDY ORIENTATION

This opening chapter has established the organization for the study. The background to the research, problem statement, aims of the research, theoretical backgrounds, research design, research method, and chapter division have all been discussed. Chapter 2 will present a thorough assessment of the literature on the dissemination and risk factors of CL in Ethiopia.

CHAPTER 2

LITERATURE REVIEW

2.1. INTRODUCTION

Contextual information, problem statement, research goal, and objectives were described in Chapter 1. The previous chapter also provides details of ethical considerations, key concepts, and highlights of the research approach. This chapter focus on a review of the works focusing on various aspects of leishmaniasis spread worldwide in general and CL in Ethiopia in particular.

The integrated literature review was utilised in this chapter with the goal of gathering and synthesise literature in a systematic manner. An integrated literature review is a method for discovering new information about a topic by examining, analyzing, and synthesising representative literature. This is done in a holistic manner in order to produce fresh frameworks and perspectives on the subject (Richard 2016:62). Internet searching, contacting authors and book review were few of the search strategies used to gatyer litratures. The scope of the litratuire review is from explaining what leishmaniasis is to advanced diagnostic and treatment methods to global burden of the disease.

More than 158 journals (such as *PLoS Neglected Tropical Diseases*, *Parasites and Vectors*, *Infectious Diseases: Research and Treatment, Asian Pacific Journal of Tropical Biomedicine, BioMed Research International, BMC Infectious Diseases, International Journal for Parasitology: Parasites and Wildlife, International Journal of Infectious Diseases and Therapy* etc.), WHO and CDC fact sheets, 10 masters and theses, 5 books,15 online resources and national guidelines were assessed. The search was from publication in 2000 to 2021. Only 8 of these were from 2000-2010 and the rest are recent publications from 2015-2021. All litratures discussing a relevant topic of interest were included.

The literatures discuss about different themes like leishmaniasis in general and Cutaneous leishmaniasis in particular. CL is discussed in detail containing

epidemiology, clinical forms, vectors, reservoir hosts, diagnosis, treatment and preventive measures. Then the literature goes on with CL in Ethiopia were information on epidemiology, management, prevalence in the country, risk factors identified so far and KAP surveys are summarized.

2.2. LEISHMANIASIS

Diseases such as dengue, lymphatic filariasis, trachoma, and leishmaniasis are neglected diseases because they have not received as much attention as other diseases (Engels & Zhou 2019:1). Leishmanaiais is one of NTDs which primarily influences the poorest people of the world. These diseases are expected to affect one billion people worldwide, with Africa having the highest proportion of cases. While some of these disorders are fatal, others cause physical disfigurement and interfere with children's intellectual and physical development (Fitzpatrick, Nwankwo, Lenk, Vlas, & Bundy 2017).

Leishmaniasis is the third most common vector-borne disease, behind malaria and lymphatic filariasis (Inceboz 2019:1). A bite from a small (2-3mm) sandfly transmits the Leshmania parasite, which has over twenty species, to humans and other vertebrates. Nearly one billion people live in leishmanaisis-endemic areas today, putting them at risk of infection. Leishmaniasis stays to be a main health challenge in four eco-epidemiological areas of the world including the Americas, West Asia, East Africa, South-East, and Asia North Africa (WHO, 2020:265). Even if hundreds of millions are at risk, Alvar et al. (quoted in Alemayehu and Alemayehu 2017:1) found that nearly 12 million people are now infected in 98 countries, with two million additional cases expected each year. Leishmaniasis transmission can be zoonotic, meaning that animal reservoir hosts such as rodents, dogs, foxes, cats, and domestic animals are involved in the disease's transmission cycle, or anthroponotic, meaning that humans are the only source of infection for the sandfly vector (Alemayehu & Alemayehu 2017:1; Sara 2019:22).

2.2.1. Historical background

Leishmaniasis is one of the diseases recorded centuries ago and paper and archeological evidence are plenty (Akhoundi, Kuhls, Cannet, Votýpka, Marty, Delaunay & Denis Sereno 2016:2). The finding of leishmanial DNA through Paleoparasitological examination of Egyptian mummies (2050–1650 BC), a sign of leishmaniasis in the earliest Egyptian medical document dating back to 1500BC, and other evidence ceased writing on the occurrence of leishmaniasis in ancient human history (Steverding 2017:4).

Even though the parasite was discovered in the late 1800s and early 1900s in lesions of cutaneous or visceral leishmaniasis, it was Dr William Boog Leishman who identified and named the parasite in 1903. The discovery of Leishman-Donovan bodies was confirmed by an Irish physician after he looks in splenocytes from kal-azar patients and observed the organisms. Donovan then called these organisms Leishman-Donovan bodies (Steverding 2017:5-6). The spread and life cycle of the Leishmania organism had been confirmed scientifically in the mid-1900s. Then the discovery of many clinical conditions and many morphologically related species and subspecies of the protozoan followed (Bari 2012:24).

2.2.2. The Leishmania parasite

Leishmania is a unicellular, dimorphic parasitic protozoa that exists in two morphological forms in humans and animals (see Figure 2.1). In the alimentary canal of Phlebotomine sandflies (the intermediate host, or vector) and culture medium, they appear as motile, flagellated, and extracellular promastigotes, or as obligatory intracellular non-flagellate amastigotes in the phagocytic cells of mammalian (vertebrate) host macrophages (Sara 2019:21). The amastigote phase morphologically consists of a circle or oval body with a nucleus, kinetoplast, and internal flagellum with a diameter of nearly 2 to 6 μm. The promastigote has a long, slender body (about 15-30 μm by 2-3 μm), a central nucleus, a kinetoplast, and a long, free anterior flagellum (Sunter & Gull 2017:2). Leishmania species can colonise two hosts thus are called heteroxenous. As a result, they can be found in

mammalian phagocytes and the intestinal tracts of phlebotomine sandflies (Akhoundi et al. 2016:13).

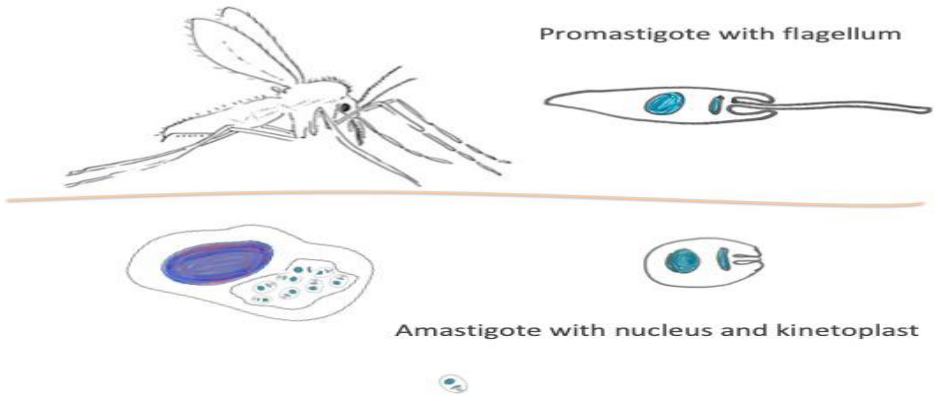


Figure 2.1: Morphological forms of Leishmania parasites (Sara 2019:21).

Various species of Leishmania have different sandfly vectors and they differ in the clinical characteristics of the disease they cause. The medically important species and their futures are shown in Table 2.1. The species recorded in Table 2.1 are splited into Old World or New World based on the geographical location. Species of the Old World include *L. tropica, L. donovani, L. major, L. infantum, and L. aethiopica.* New World species consist of *L. amazonensis, L. Mexicana, L infantum chagasi, L. brazilensis, L. panamensis, L. guyanensis, and L. peruviana.* As far as humans are considered, *L. tropica, L. major, L. donovani, L. mexicana, and L. braziliensis* are the most important species (Nuako 2016:23-24).

Table 2.1. Major diseases causing Leishmania and their future (Thomas 2018:4-6).

Species	Disease in humans	Notable futures	Geographic distribution	Mammalian host	Vectors
Leishmania (*Leishmania*) *Major*	Cutaneous (oriental sore)	Rural zoonotic	North Africa, Sahel of Africa, Central and West Asia	Great gerbil (Rhombomys opimus), fat sand rat (Psammomys obesus)	Phlebotomus papatasi, P dubosqi, P salehi
L. tropica	Cutaneous (oriental sore)	Urban anthroponotic	Central and West Asia	Humans	Psergenti
L. aethiopica	Cutaneous diffuse cutaneous	Rural zoonotic	Ethiopia, Kenya	Rock hyraxes Heterohyrax brucei (Procavia spp.)	P longipes, P pedifer
L. donovani	Visceral (kala-azar)	Epidemic anthroponotic	The Indian subcontinent, East Africa	Humans	P argentipes, P orientalis, P martini
L. infantum	Infantile visceral	Zoonotic peridomestic	Mediterranean basin, Central and West Asia	Domestic dog	P ariasi, P perniciosus
L. mexicana	Cutaneous (chiclero ulcer)	Sylvatic zoonotic	Central America	Forest rodents (*Ototylomys phyllotis* and others)	Lutzomyia olmeca olmeca
L. amazonensis	Cutaneous	Sylvatic zoonotic	South America	Forest rodents (*Proechimys*spp. and others)	L flaviscutellata
Leishmania (Viannia) braziliensis	Cutaneous mucocutaneous (espundia)	Sylvatic zoonotic	Central and South America	Forest rodents (*Akodon* spp., *Proechimys* spp. and others)	L wellcomei, L complexus, L carrerai
L. peruviana	Cutaneous (uta)	Upland zoonotic	Peru	Reservoir unknown, dog	L peruensis, L verrucarum
L. guyanensis	Cutaneous, often metastatic (pian-bois)	Sylvatic zoonotic	South America	Sloth (*Choloepus didactylus*), anteater (*Tamandua tetradactyla*)	L umbratilis
L. panamensis	Cutaneous	Sylvatic zoonotic	Central America	Sloth (*Choloepus hoffmanni*)	L trapidoi

2.2.3. The parasite life cycle

The sandfly stage, which is a flagellated promostigote stage, and the human/animal stage, which is a non-flagellated amastigote stage, are the two stages of leishmania's natural life cycle (Figure 2.2).

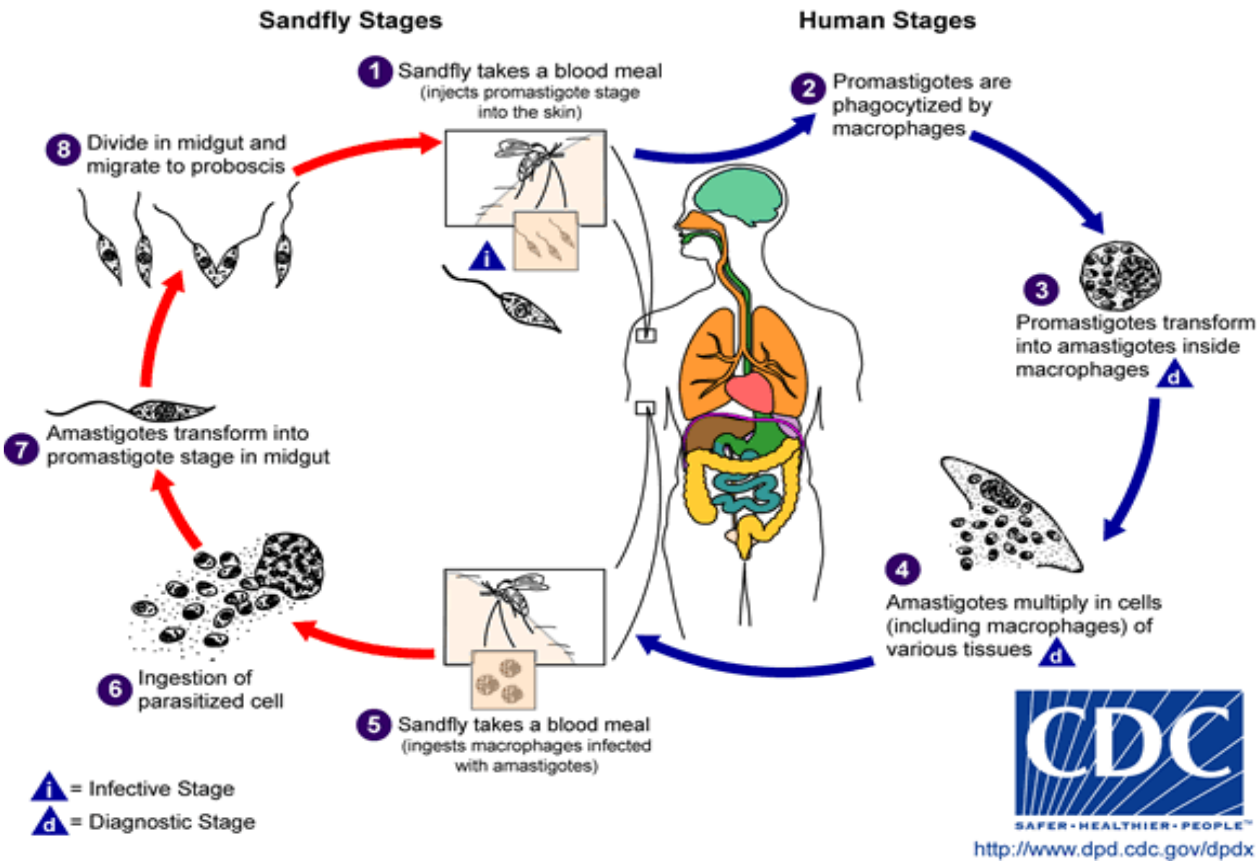


Figure 2.2: The life cycle of Leishmania (CDC:2020).

The infective promastigotes stages of Leishmania are injected by infected female phlebotomine sandflies into the skin from their proboscis during blood meals ❶. Then macrophages phagocytise promastigotes❷. Promastigotes transform into amastigotes in these cells❸,which in turn proliferate by simple division and become infective to other mononuclear phagocytic cells❹. Disease development is dependant on Parasite, host and other factors. When Sandflies ingest affected cells during blood meals, they become infected (❺, ❻). In the gut of Sandflies amastigotes is transfomed into promastigotes ❼ and migrate to the proboscis❽.

Human phase: During a blood meal, contaminated sandfly infuses promostigotes into the skin which are phagocytized by neutrophils circulating at the bite spot. Neutrophils distribute the promostigotes, which are then swallowed by various defense cells in various organs, where they transform into amastigotes, the parasite's tissue stage. Amastigote multiplies by simple division in the cell (including macrophage) of different tissues and damage of host cell triggers the spreading of the parasite into blood and lymph system and the parasite are once again engulfed by new cells (Maza, 2014:16; CDC, 2020).

Sandfly phase: while taking another blood meal, sandfly swallows affected macrophages holding amastigotes from infected persons then amastigotes roam in the midgut of sandfly and convert into promastigotes which after multiplication reach to anterior midgut and foregut. If the contaminated sandfly feeds on blood and injects promastigotes into the skin of a mammalian host, a new human stage will begin (Sunter & Gull 2017:2; CDC 2020).

The disease progresses in a variety of ways, from skin ulcers (CL) to a fatal circulatory condition (Visceral Leishmaniasis, VL), which is usually fatal due to the lack of therapy (Sunter & Gull 2017:2; CDC 2020).

2.2.4. Clinical forms of leishmaniasis

A variety of clinical characteristics make leishmaniasis a complicated disease, ranging from mild skin lesions to fatal visceral manifestations. In general, the disease's clinical presentation and progression are polymorphic. Clinical presentation may differ due to Leishmania species or strains, host genetic underpinnings, geographical location, and immunological response (Torres-Guerrero, Quintanilla-Cedillo, Ruiz-Esmenjaud & Roberto Arenas 2019:7). Visceral leishmaniasis (VL) or kala-azar (fatal without treatment) and Cutaneous Leishmaniasis (CL), a scarring and stigmatizing disease, are the two main clinical forms of the disease (which can take the form of Localized CL, MucoCL, Disseminated CL and recidivans) (Siriwardana, Deepachandi, Gunasekara, Warnasooriya, & Karunaweera 2019:1 and Machado, Prates & Machado 2019:10). CL and its four clinical forms are to be discussed later.

As shown in Figure 2.3, Visceral Leishmaniasis (VL), often known as kala-azar, affects multiple internal organs (most commonly the liver, spleen, and bone marrow). In Asia and Eastern Africa, *L. donovani* causes the disease, while *L. infantum* causes it in Latin America, Central Asia, and the Mediterranean region (Lindoso, Moreira, Cunha & Queiroz 2018:193). There are two forms of VL based on the susceptible species. Anthropogenic visceral leishmaniasis (AVL), which is

shared between humans by vector carriers throughout Asia and Africa, is known as anthroponotic visceral leishmaniasis (AVL). Zoonotic visceral leishmaniasis (ZVL) is a disease that is transmitted from humans to animals (such as dogs and rodents). In Latin America and the Mediterranean region, the main parasites responsible for ZVL are *L. donovani*, *L. infantum*, and *L. archibaldi* (Alves, Bilbe, Blesson, et al. 2018:2).

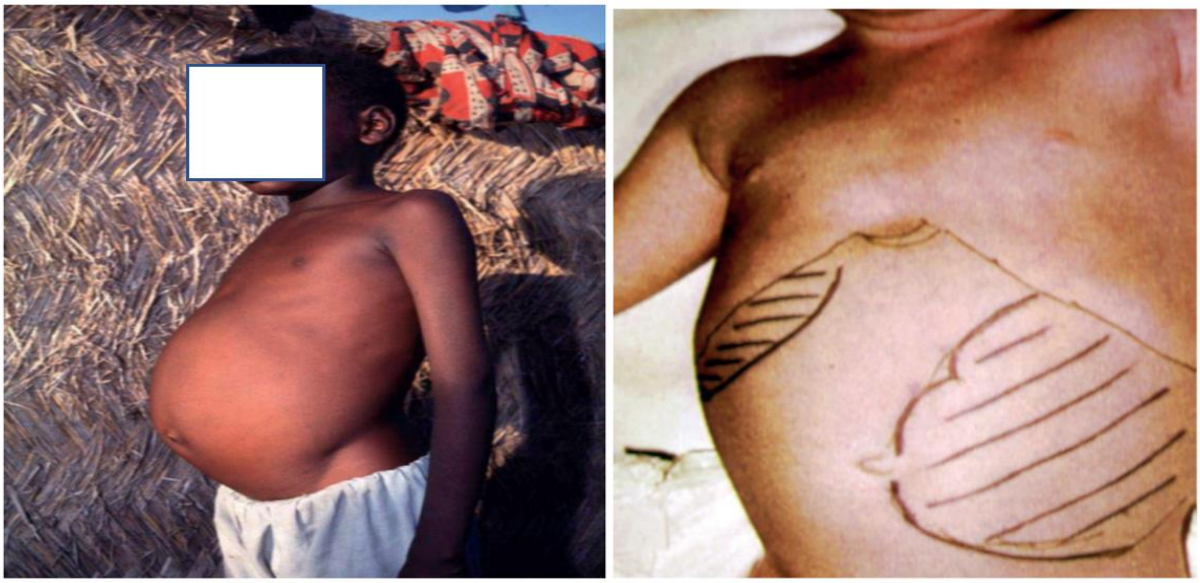


Figure 2.3: Visceral leishmaniasis in children with hepato-splenomegaly.
The size of the liver and spleen are depicted on the skin in the right image, and the spleen is extremely inflated (WHO, 2016).

Whether zoonotic or anthroponotic, all forms of leishmaniasis transmit through the bite of female sandfly vectors. *L. infantum* is transmitted by 31 different Sandfly species, while *L. donovani* is transmitted by 9 different Sandfly species. The principal vectors are sandflies (*Phlebotomus* spp. (Old World) or *Lutzomyia* spp. (New World), which are small insects with hairy exoskeletons. Their eyes are huge and black, and their legs are lengthy and stilt-like. Sandflies have a fourteen-day lifespan on average. Sandflies are most active from dusk to dawn, including the hours of twilight, evening, and night. Female sandflies need proteins to produce eggs thus drink blood while plant sugars are food source of male sandflies (Rutte 2018:10).

VL is more difficult to detect and treat than other kinds of leishmaniasis because it affects the internal organs. Organomegaly (e.g., huge splenomegaly and modest hepatomegaly) (Figure 2.3), fever with distinct patterns of rising 2-4 times/day, wasting and weight loss, deficiency of all three cellular components of the blood, and elevated levels of gamma globulin are five distinct features of VL caused by all species. Generally, the extremely popular clinical elements are fever, anemia and splenomegaly. VL commencement can be undetected or acute. Like in CL, skin signs in VL are common. VL is also known as "Kala-azar," which means "black disease" and refers to the earth-gray skin color seen in sick patients, particularly in India (Karimi, Alborzi & Amanati 2016:4). If left untreated, VL, the world's most dangerous parasite illness after malaria, is almost invariably fatal (Global Burden of Disease 2015).

After VL is treated, a condition known as post kala-azar dermal leishmaniasis (PKDL) may occur (Figure 2.4). PKDL is a skin illness that manifests itself in a variety of ways, ranging from a simple hypopigmented macular form to more advanced lesions such as papular or nodular lesions on the face, trunk, and upper arms (WHO 2012).

Figure 2.4: Post kala-azar dermal leishmaniasis (PKDL) (WHO 2012).

PKDL is restricted in South Asia (India, Bangladesh and Nepal) and East Africa (mainly in Sudan) because PKDL is primarily confined to follow VL due to *L. donovani*. In Sudan and India, PKDL occur after cured VL in 50% and 5–10% case, respectively (Rutte 2018:13). Risk factors for PKDL are not well known. Previous

treatment of VL with inadequate dosage of drug and the drug used, malnutrition, HIV infection, and young age may play a role (Gedda, Singh, Kumar, et al. 2020:1). The social stigma associated with PKDL is the most significant impact, especially if it develops on the face while the disease is not life-threatening. Another important concern with PKDL is the fact that the lesion is rich in parasite, and this may serve as a source of contamination for other individuals while sandflies are taking blood meal. Eradication of PKDL should be included as a part of the VL elimination programme (Gedda, Singh, Kumar, et al. 2020:1).

According to the WHO, VL causes 200,000–400,000 episodes per year, with 20,000–40,000 deaths. According to the World Health Organization, Sudan, Ethiopia, South Sudan, Brazil, Somalia, India, and Kenya account for more than 90% of all new VL cases (WHO, 2018). South Sudan, Ethiopia, and Sudan account for the largest percentage of new cases in East African countries, which are the second most important VL focus after the Indian subcontinent, with 30,000 to 40,000 new cases annually. In Ethiopia, the annual incidence of infections ranges from 2500 to 4000, putting approximately 3.2 million people at risk of infection (Gadisa, Tsegaw, Abera, et al. 2015:1). The status of the endemicity of VL worldwide is shown below in Figure 2.5.

For the diagnosis of VL, different methods are applicable such as parasitological diagnosis, molecular diagnosis, and serological diagnosis. Parasitological diagnosis continues to be the gold standard since it is highly specific. The amastigote forms are visible under the microscope in tissue samples taken from aspirates of bone marrow, lymph nodes, or spleen during parasitological examination (Rutte 2018:16). To detect parasite DNA in the blood, molecular techniques such as polymerase chain reaction (PCR) and real-time quantitative PCR (qPCR) are utilised. Diverse serological tests exist to detect human antibodies against the parasite, including the direct agglutination test (DAT), enzyme-linked immunosorbent assay (ELISA), indirect fluorescent antibody test (IFAT), and rapid immunochromatographic assays, such as rK39, which is available as a dipstick. Another important serological test is use of leishmanin skin test (LST) which can detect past exposure and is particularly

important to asesse previous exposure of the community to the disease (Rutte 2018:16).

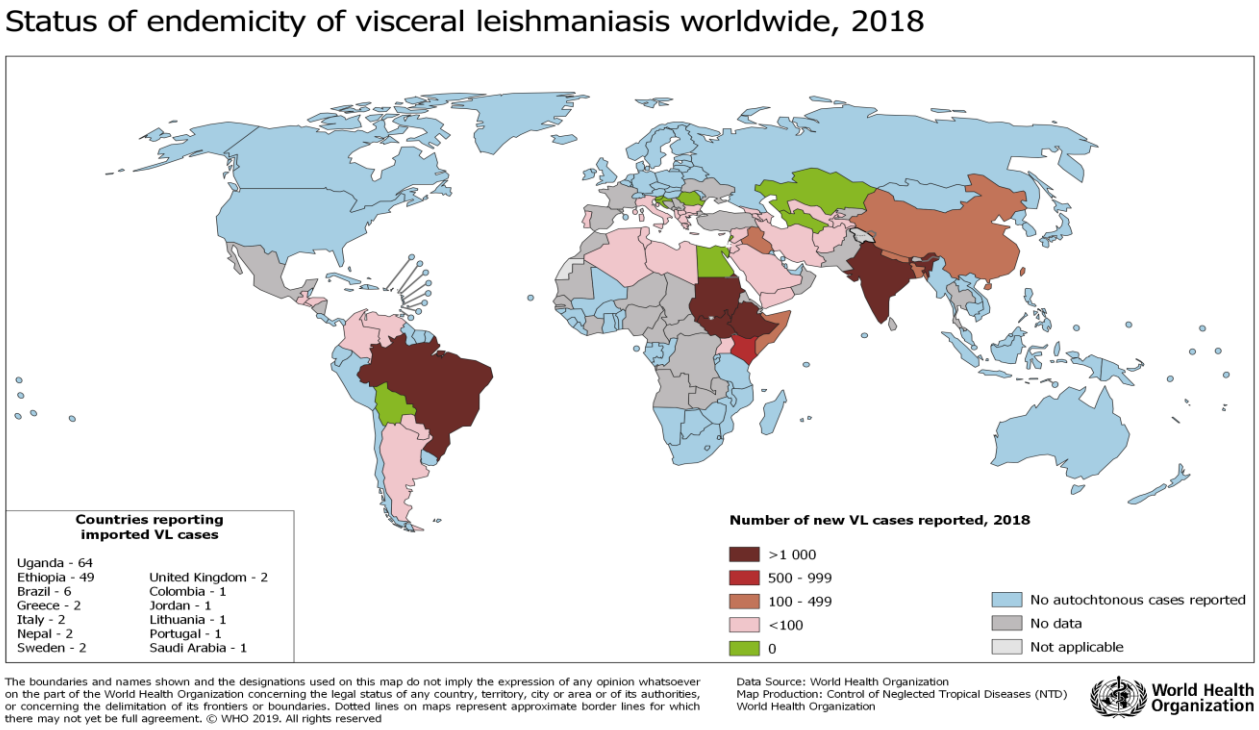


Figure 2.5: Status of endemicity of VL worldwide (WHO 2020).

Treatment of VL depends on the geographic region and the existence of a cold-chain and parasite resistance. Treatment involves use of antileishmanial therapy. The key limitations on the choice of antileishmanial drugs are cost and accessibility (WHO, 2012). These medicines are given intramuscularly/intravenously for 30 days posing treatment defaulting, parasite resistance, infection setback and development of PKDL. Recently, liposomal amphotericin B, amphotericin B, paromomycin, and miltefosine have been approved for the treatment of VL in a number of countries (WHO, 2012). In general, leishmaniasis management focuses on reservoir and sandfly control, early diagnosis, and adequate treatment. Controlling anthroponotic visceral leishmaniasis (AVL) and zoonotic visceral leishmaniasis (ZVL) requires distinct techniques (ZVL). Because there is no effective human vaccine, AVL prevention relies primarily on early detection and treatment, or the use of long-term pesticide nets. Nonetheless, ZVL, because dogs are the most prevalent mammal carriers of the virus. ZVL control methods include dog immunizations, dog culling,

and the use of pesticide collars, which are all more frequent than AVL control methods (Bi, Chen, Zhao, Kuang, & Wu 2018:1).

Co-infection with HIV is an issue with VL control. Around the world, the AIDS pandemic has changed the epidemiology and natural history of leishmaniasis (Nuako 2016:30). Co-infection with HIV, which occure in 22.2% of all VL cases, is a double burden because both illnesses equally boostone another in such a way that HIV increase the likelthood of VL infection and VL facilitates for HIV multipilication (Zijlstra 2014:2). Presently, Ethiopia is the sole possessor of the major proportion (40%) of this type of co-infection (Diro, Lynen, Ritmeijer, Boelaert, Hailu, Griensven 2014:2). However, countries like Asia and Latin America have few records (Zijlstra 2014:2).

Since this study focuses on CL, a detailed literature review of CL is presented next.

2.3. CUTANEOUS LEISHMANIASIS

This from of the disease is known to have the highest prevalence in the world. As a result, CL was established as a crucial neglected tropical disease by WHO (Sakhaei, Darrudi, Motaarefi & Sadagheyani 2019:3710). CL begins with erythematous papules, then developes to nodules and lastly, to crusted ulcerated lesions at the bite site because of replication of parasites in the dermis (Nuako 2016:24). Therefore, CL produces skin lesions or ulcers on uncovered parts resulting in permanent scars and serious disability or stigma (WHO, 2020). Although CL is not deadly, its impact on the local communities is devastating. As an example, it can cause skin lesions that can leave permanent scars. The scars are source of mental, social, and emotional torments for affected individuals especially children and women (Hussein, Balatay, Saleem, et al. 2019:2).

CL can be classified based on different factors such as mode of transmission and geographic location. If mode of transmission is considered there are two types of CL namely zoonotic and anthroponotic CL and based on geographical location as Old-World CL (oriental sore) and new world CL. Old World CL is mainly found in Iran,

Afghanistan, the Middle East, India, south Russia, southern Europe North, and East Africa. *L. major*, *L. tropica*, and *L. aethiopica*, as well as *L. infantum* and *L. donovani*, are the causal agents. In the New World of Central America and Mexico, *Leishmania braziliensis* and *Leishmania mexicana* are aiteologic agents (Tamiru et al. 2019:3). The *Phlebotomus* species transmit the Old-World CL, while the *Lutzomyia* species transmit the latter (Ghatee, Taylor & Karamian 2019:1).

2.3.1. Epidemiology of CL

As mentioned before, lesihmanaisis has complicated epidemiology ownig to moret than twenty pathogenic species of Leishmanais and presence of more than 30 preven sandfly vector for humansthe (Degu 2006:3). Each *Leishmania* species has intimate connection with reservoir host, snadlfy vector and its biotope in each geographical area (Degu 2006:3).

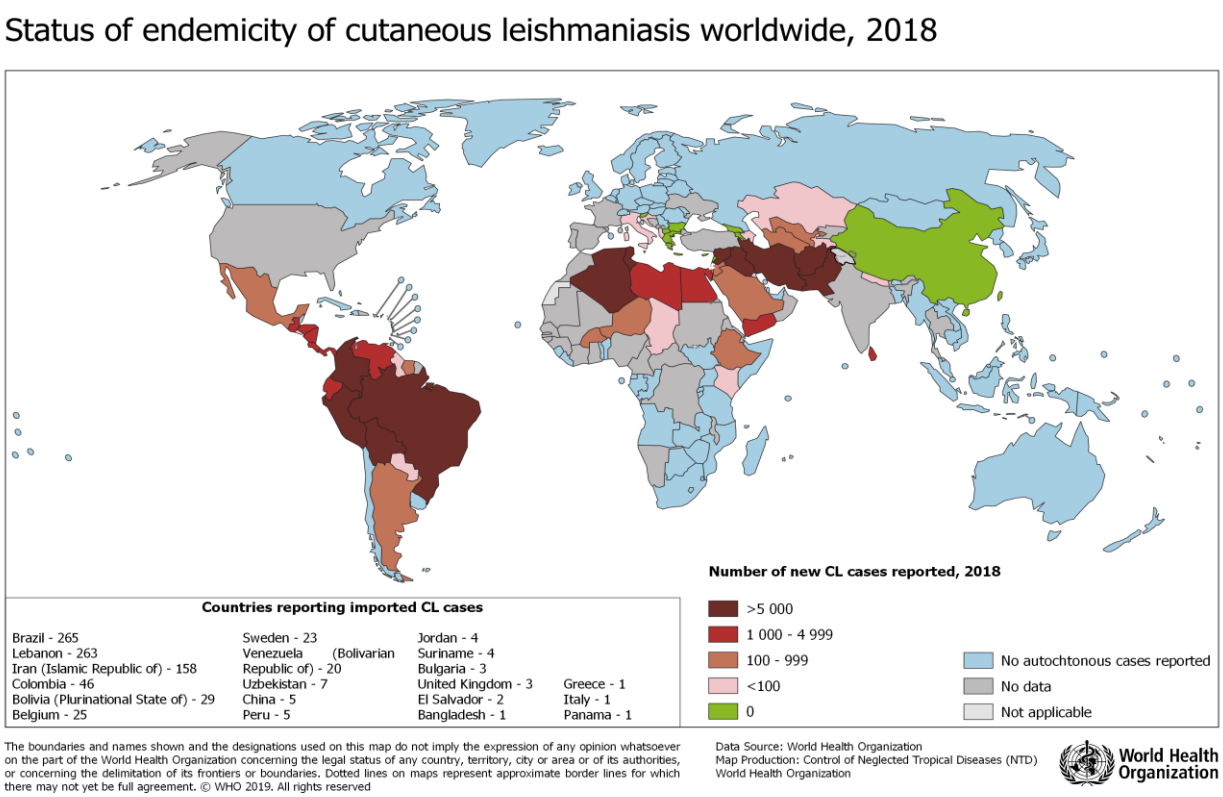


Figure 2.6: Status of endemicity of CL worldwide (WHO 2020).

Leishmaniasis is widespread disease occurring in 88 countries with 1.5 million incidence of CL per year. But according to WHO, the Some countries such as

Algeria, Colombia, Brazil, Afghanistan, Iran, Sudan, Ethiopia, Peru and Syria Nicaragua have profound spread as they carry 70% of incidence (WHO 2010). The status of endemicity of CL worldwide is shown in Figure 2.6 above.

Economic development and behavioural and environmental changes e.g., new resettlements, invasion into primary abandoned forsets, desertification, rural to urban immigration, fast and spontaneous urbanisation, construction of dams, new irrigation structures can all casuse increase in intensity of contact with sandfly vectors. As a result of these factors, there has been a noticeable and concerning increase in the number of CL cases in various parts of the world. CL in Brazil, where cases increased from 21 800 in 1998 to 35 000 in 2000, CL in Kabul, Afghanistan, where cases increased from 14200 in 1994 to 200 000 in 1999, and CL in Aleppo, Syria, where additional 2000 cases were recorded between 1998 and 2000. People living in newly built residential areas have highest risk of infection due to sandfly preference to live near these residents looking for suitable host (Pigott, Bhatt, Golding, et al. 2014:2). Considering its medical burden and mortality in war-torn countries, CL is believed to be among the top nine infectious diseases that inflict significant health problems (Jorjani, Mirkarimi, Charkazi, et al. 2018:2).

2.3.2. Clinical forms of CL

Although the genus Leishmania contains a large number of species, CL is caused by just roughly 20 of them (Tamiru et al. 2019:3). In the Old World, *Leishmania tropica*, *Leishmania major*, and *Leishmania aethiopica* are the most common aetiologic agents (Nuako 2016: 25, Sara 2019:25). For CL, there is a broad range of clinical demonstrations. This may be attributed to variability in parasite species, strain, virulence, and other intrinsic characteristics and variability in the host genetic factor and immune response (Sara 2019:25). The cutaneous syndromes associated with CL *Leishmania* parasite infection include Local CL (LCL), Leishmaniasis Recidivans (LR), Diffused CL (DCL), and MucoCL (ML) (Sara 2019:25).

2.3.2.1. Localised CL

L. major, *L. tropica*, *L. braziliensis*, and *L. Mexicana* cause localized CL (LCL) in both the Old and New Worlds. The most common clinical symptom of leishmaniasis is LCL. After one week to three months of an incubation period, the LCL lesion started to show asa papule red in color and then expands to become a plaque with nodule which finaly grows as ulcer characterised by circumscribed and violaceous borderline and this also contain crusted hypertrophic margins which is considered to be pocket of the parasite (Nuako 2016: 26) (Figure 2.7). In uncomplicated case of LCL, parasites persist localised in skin tissue and result in chronic ulcer of the skin which heal slowly (Nuako 2016: 26). Even if lesion of LCL is not painfull and heal over time with out medication, the healing process take months and sometimes years and it also leave behind disfiguring scars. Additionally, lesions can become complicated by secondary bacterial or fungal infection complicating diagnosis and treatment (Scorza, Carvalho & Wilson 2017:2).

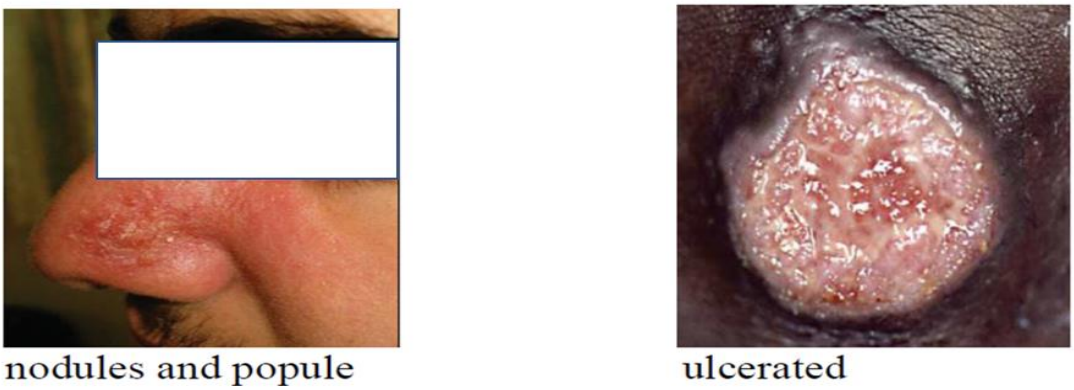


Figure 2.7: Developmental stages of Localized CL (Nuako 2016:25).

2.3.2.2. Leishmaniasis Recidivans

Leishmaniasis recidivans is another rare clinical type of leishmaniasis. After the original infection has cleared, this manifests as a recurrence of lesions (Figure 2.8) (Stark 2020). The factors that cause the disease to relapse are being researched. Leishmaniasis recidivans is caused mostly by *L (Viannia) braziliensis* in the Old World (Mediterranean, Middle East, India, and China) and *L tropica* in the New World (Mediterranean, Middle East, India, and China) (Stark 2020).

Figure 2.8. Leishmaniasis recidivans (left: -Bari 2012 and right: Dassoni et al. 2017:107).

2.3.2.3. Diffused CL

Diffuse CL (DCL) is characterized by widely widespread cutaneous macules, nodules, papules, or plaques, as well as dispersed penetration on the limbs and face, and is disseminated mostly by *L. aethiopica* (See Figure 2.9.). Unlike LCL, DCL does not ulcerate and not heal naturally, and may relapses after cure. In sub-Saharan countries, nearly all cases are registered from Ethiopia (Gyapong & Boatin 2016:101).

.

Figure 2.9: Clinical presentation of Diffused CL (left-Torres-Guerrero et al 2017:9; right- Gyapong & Boatin 2016:102).

L aethiopica causes DCL in Old World countries like Ethiopia and Kenya. The causal agents of DCL in the New World differ depending on where they are found. *L mexicana* is found in southern, central, and northern America, *L amazonensis* is found in the Dominican Republic and Central and South America, and *L venezuelensis* is found in Venezuela (Stark 2020).

2.3.2.4. Muco-CL

In some cases, after months and even years of recovery of CL, remaining parasites travel to nose parts and produce mucoCL (MCL) which is also known as Espundia (Sara 2019:27) (Figure 2.10). The starting point of lesion development is the boundary of skin and mucosa which then expands to significant damaging lesions. The lesions will progress causing deterioration of mouth and nose tissues. This may ultimately lead to eating difficulties and breathing (Sara 2019:27). If a person exhibits the above clinical indications and parasite presence is confirmed by parasitological or serological tests, the person is diagnosed with MCL, according to WHO classification (WHO 2010).

Figure 2.10: Presentation of MucoCL (Bari 2012).

L. aethiopica, like DCL, is the Old-World cause of MCL in Ethiopia, Kenya, and Namibia. MCL is caused by Leishmania species such as *L. braziliensis* and *L. panamensis* in New World nations such as Central and South America. MCL is caused by *L. guyanensis*, *L. mexicana*, and *L. amazonensis* in nations such as America, Brazil, Guyana, Panama, and Surinam (Stark 2020).

2.3.3. Vector of CL

CL is a vector-borne infection transferred when infected female bites a host. Sandfly adults are golden, brownish, or gray coloured tiny flies with a length of 3 milimeters. The mouthparts are long and piercing and are well modified for drawing blood from their carefully chosen host (European Centre for Disease Prevention and Control 2020). Distinguishable characteristics that help identify sandflies from some other small flies is their resting position where they hold their hairy-looking wings in a vertical V-shape above the abdomen. Sandfiles can also be identified by their extremely long six legs on the adults which are even longer than the entire body, they have hairy covering, and they tend to hopp around the host before biting (European Centre for Disease Prevention and Control 2020). The structure of the vector is shown in Figure 2.11 below.

For general activities such as fighting and flight, both males and females consume sugars from aphid honeydew or directly from plants (Adam, Hassan, Abdelnour & Awadallah 2017:16). Only females suck blood because they want blood for the development of their eggs. Vertebrates such as amphibians, birds, reptiles, and mammals are target hosts (Tanure, Peixoto, Afonso, Duarte, Pinheiro, Coelho & Barata 2015:321).

Figure 2.11. Female sandfly, *Phlebotomus papatasi* taking blood meal (Nuako 2016:46).

Sandfly larvae feed on organic waste, hence sandfly breeding sites are typically damp spots such as soil fissures, animal burrows or caves, wall and rock crevices, and damp leaf debris in forests with ideal temperature and moisture (Adam et al. 2017:15&16). In general, sandfly bites are very painful. Unless disturbed while resting, sandflies bite humans at throughout the evening and the night. They need dark humid places to rest during the daytime and can be seen in human residences between cloths, behind closets, hanging pictures and in the wall crevices. Caves, rock crevices, soil cracks and fractures, stream banks, animal burrows, and leaf litter in forests are examples of humid microhabitats where wild species can be found (Adam et al. 2017:15&16).

Sandflies are from the Diptera (order), Nematocera (suborder), Psychodidae (family), and Phlebotominae (subfamily). *Phlebotomus*, *Sergentomyia*, and *Chinius* are Old World genera, while *Lutzomyia*, *Warileya*, and *Brumptomyia* are New World genera. So far, roughly 800 sandfly species have been identified, with 464 in the New World and the rest in the Old World (Akhoundi et al. 2016). The only established vectors of human leishmaniasis are species and subspecies of the genera *Phlebotomus* in the Old World and *Lutzomyia* in the New World, so none of these species are important for leishmaniasis transmission. The very species-sandfly specific nature in the transmission of CL is that one species of the parasite is carred or transmitted by one sandfly species and each Leishmania species cause a unique type of CL (Alemayehu & Alemayehu 2017:2).

2.3.4. Reservoir hosts of CL

A reservoir host is any person, animal, plant, soil, or substance which is important for the survival and reproduction of an infectious agent in such a way that it can be spread to a vulnerable host (Aryal 2019).

Animal reservoirs can serve as a means by which Leishmania species are preserved as they are essential for spread of zoonotic and rural or sylvatic CL. Sylvatic cycle is when a part of the parasite's life cycle is circulating among wild

animals and vectors (Alemayehu & Alemayehu 2017:4). Humans become incidental or dead-end hosts when they break in the sylvatic cycle and infected by a vector (Alemayehu & Alemayehu 2017:4). The Leishmania parasite has been found in a variety of wild and domestic mammal species all over the world. Rock hyraxes, mongooses, rodents, dogs, foxes, cats, wolves, jackals, monkeys, armadillos, bats, and other domestic animals are just a few of the identified reservoirs for Leishmania in different places (Rohousova, Talmi-Frank, Kostalova, et al. 2015). Owing to the complex nature of Leishmania reservoirs, identification of an animal as Leishmania reservoir can be attained by local studies focusing on the ecology of CL transmission (Roque & Jansen 2014:253). Diverse types of reservoir host important in the maintenance of the Leishmania parasite are listed in Table 2.2.

Table 2.2. Type of Leishmania reservoir hosts in different countries (Alemayehu & Alemayehu 2017:4).

Region	**Countries**	**Reservoir hosts**
Old world	North Africa, central and west Asia	Dog, human, rodent
	Ethiopia, Kenya	Rodents, dog, domestic animals, bats, human, rock hyrax
	The Indian subcontinent, (India, Nepal, Bangladesh) and east Africa	Dog, human, rock hyrax, rodent
	Mediterranean basin, central, west Asia and West Africa	Dog, fox, rodent, human
	Europe	Dog, fox
New world	Argentina, Belize, Bolivia, Brazil, Colombia, Costa Rica, Dominican, Ecuador, El Salvador, French Guyana, Guadeloupe, Guatemala, Guyana, Honduras, Martinique, Mexico, Nicaragua, USA, Venezuela, Paraguay, Peru, Surinam, Panama,	Dog, cats, rodent, marsupials, anteater, fox, monkey, coati, sloth, armadillo, porcupines, kinkajou, raccoon, red squirrel,

To be called a reservoir, a host must transmit the parasite to the sandfly vector. The ability of a reservoir host to support the formation of sandflies is thus an essential condition for any mammal to be termed a Leishmania reservoir (Akhoundi 2016). The ability of the reservoir host to be a competent transmitter of the disease and availability of Leishmania to the vector are key components for the maintenance of leishmaniasis a specific area (Akhoundi 2016).

2.3.5. Diagnosis of CL

Because of the varied symptoms ranging from skin lesion to disminated mucosal infection and the unique species of Leishmania involved, the diagnosis of CL is difficult. Confirmatory diagnosis requires Leishmania species specific laboratory methods as VL, CL, or MCL exibit unique symptoms. Methods used for the diagnosis of CL are microscopy, culturing, molecular methods, serology, and other methods (Sara 2019:28).

2.3.5.1. Microscopy

Since the Leishmania parasite have been discovered light microscopic examination of aspirates or biopsy samples has been the major confirmatory test for leishmaniasis. In this test microscopic slides are stained with Giemsa stain to observe the round or avoid amastigotes (Leishman-Donovan bodies). For confirmation, the nucleus and a kinetoplast are apprciated within tissue macrophages (Sara 2019:28). Depending on the type of disease, samples for microscopic tests can be collected from various parts of the patient's body, including bone marrow aspirates, lesion scrapings, spleen aspirates, lesion aspirates, and biopsies (Sara 2019:28). Examination of amastigotes in the smear is the criterion of CL diagnostic. Even though this method is cheap, accessible, and highly specific, its sensitivity is low for all tissues (53-86%) (Rasti, Ghorbanzadeh, Kheirandish, et al.2016:611).

2.3.5.2. Culturing

Novy-MCNeal-Nicolle-medium or substituted liquid cell culture medium can be used for culturing of Leishmania parasites and to grow the promastigote forms (Sara 2019:28). These mediums are specialy prepared to culture amastigote invitro and isolate promastigotes. Though these are valuable tool, the demand of advanced laboratories, elevated risk of contamination and time taking procedures are few of the shortcomings. Since culturing is not able to identify parasite species causing the disease, molecular methods are needed (Nuako 2016:41).

2.3.5.3. Molecular methods

Molecular metods are used recently even to detect small amounts of parasite DNA but they can only be performed in a setting with high resources. Microscopy and culture approaches have been shown to be less sensitive than molecular techniques. Leishmania parasite species can also be identified using molecular techniques (Nuako 2016:42). Different molecular approaches offer varying degrees of sensitivity and precision, but quantitative PCR-methods are the most sensitive. PCR is currently the method of choice for species identification. Sequencing or gel analysis of amplified genes such as HSP70 or ITS1 is part of the PCR process. PCR can be done on samples form skin or tissue or from culture strains (Sara 2019:29). PCR is a rapd, overly sensitive, highly specific and multipurpose method which makes it the most preferred method among others (Nuako 2016:42).

2.3.5.4. Serology

There are a variety of serological (anti-Leishmania antibodies in peripheral blood) tests available on the market, including immunofluorescence assays (IFATs), which require an immunofluorescence microscope, and enzyme-linked immunoassays (ELISAs), which do not (Sara 2019:30). These procedures necessitate the use of specialised equipment to read the results, making them more appropriate for advanced and resourceful diagnostic research laboratories. Direct agglutination assay (DAT) and immunochromatographic diagnostic assays, which can identify antibodies generated against the leishmanial antigen, are the most commonly used serological diagnostics in endemic areas with limited resources. These methods are rapid are used and are read optically without equipment (Sara 2019:30).

2.3.5.5. Choice of diagnostic method for CL

The choice of diagnostic methods varies depending on the purpose of the diagnosis. For example, in day-to-day diagnosis of CL, mainly direct microscopic test and culture tehcniques are used. Whereas molecular approaches are mostly used for research purposes (Ertabaklar, Çalışkan, Boduç & Ertuğ 2015:1). The availability of resources, time and laboratory setup are other determinants for the choice of diagnostic tests. Any disgnostic method must have high sensitivity and be

specific to avoid devastating effects in the case of false negative and false positives (Sara 2019:31).

Till now there is no single diagnostic technique that is accepted as a gold standard for detecting leishmaniasis. So far, the best leishmaniasis diagnostic method is the use of molecular techniques such as PCR which provide significant benefits in specimen gathering, transport and DNA extraction. Various investigators have stated that PCR has 100% specificity with sensitivity ranging between 92 and 98% (Rasti et al. 2016:613).

In a study comparing diagnostic methods for leishmaniasis, positivity rates for direct microscopic examination, 92.0% for culturing, and 81.1% for molecular methods in CL positive patients were 78.4% for direct microscopic examination, 92.0% for culturing, and 81.1% for molecular methods (Ertabaklar, alşkan, Boduç & Ertu 2015:1). Another molecular technique was shown to be the most successful in a comparable comparison investigation (Rasti et al. 2016:613). According to the findings of the aforementioned research, using more than one technique for diagnosing CL improves sensitivity and specificity, making it easier to diagnose a wide variety of patients (Rasti et al. 2016:613).

2.3.6. Treatment of CL

Treatment guidelines for leishmaniasis are moving in the direction of a species identification methodologies through molecular approaches. Consequently, detection of different species of the *Leishmania* parasite is essential to assist proper treatment (Showler & Boggild, 2015:1). However, identifying the Leishmania species is not sufficient for treatment as thoughts have to be made about other burning issues like the risk MCL development after CL and the rsiak of PKDL development after cure of VL. The immunity status of the host, toxicity of treatment, preference of the patient and their commitment to complete the therapy are other factors of concern (Showler & Boggild, 2015:1). To date, vaccines are not available to effectively prevent leishmaniasis in humans plus guidelines for prophylactic treatment are scarce. Treatment options following confirmatory disagnosis of Cl are

many. In addition to the possibility of having potentially serious side effects owing to accumulation in the tissues, systemic drugs are not accessible to people in developing countries owing to infrastructure or cost (Sara 2019:31).

Unless it is MCL which can result in incapacitating and dreadful destruction of the affected tissue, CL typically heal by itself. During the healing process which can take months and even years, the infected person is expsed to functional impairment, secondary bacterial or fungal infection and development of marring scar which is permanent. If evidence is scarce such as identification of Leishmania species is unattainable treatment is mainly dependant on opinions of local experts (Vries Reedijk & Schallig 2015:104). CL treatment can be done using topical medication or systemic therapies. The choise between these two depend on many factors including species of Leishmania, geographic areas, existence of immunosuppression, duration of therapy, size, and location of lesions and clinical manifestations (Gradoni, Lopez-Velez & Mourad 2017:29-31). Thermotherapy, cryotherapy, paromomycin ointment, and local antimonial infiltration may be promising choices for topical treatment with little blood toxicity. Medicines used for systemic treatment include azole drugs, miltefosine, pentavalent antimonials, pentamidine, and amphotericin B and its liposomal version (Alidadi & Oryan 2014:1). There is no ideal drug for treating the disease. Available drugs are toxic, take long duration, cause harmful reactions which may force patients to stop treatment. Researchers also indicated that currently used drugs are unable to completely remove the parasite from infected persons (Menezes, Guedes, Petersen, Fraga & Veras 2015:1).

Dose reduction, shortage of treatment duration, avoidance of adverse reactions and provision of efficacy and patient safety was achived through use of alternative protocols. These include rational combination of drugs/immunotherapy and controlled release systems (like liposomes and nanoparticles). Yet, confirmation on the effectiveness of these protocols needs comprehensive long-term investigations (Menezes et al. 2015:7).

2.3.7. Preventive measures and control of CL

For both the patient and the general public, "prevention is better than cure," hence prevention is an important tool in the control of CL (Maza 2014:22). However, vector-borne parasitic diseases are epidemiologically complex with sophisticated and diverse parasite-vector-host interactions. Epidemiological studies of sandfly-*Leishmania*-host interactions explaining the disease transmission are often difficult to obtain in NTDs. Understanding disease transmission is crucial to be able to prevent and control the disease plus control measures need to be cost-effective and environmentally sustainable (Maza 2014:22).

According to World Health Organization, early diagnosis, integrated vector management, control of animal reservoir hosts, efficient disease surveillance, successful timely treatment, reinforcing partnerships and social enrolment are directions given for control of CL in humans (WHO, 2020).

2.3.7.1. *Early diagnosis and treatment of cases*

Early identification followed by effective prompt treatment can help prevent disabilities and death due to the disease. Setting disease detection mechniams can help decrease spread, examine, and measure the disease (WHO, 2020).

2.3.7.2. Integrated vector management

Reduction of sandfly numbers by application of insecticide, use of insecticide impregnated bed nets, keeping environmental hygine, and applying personal measures is vital for decreasing or intereption of disease spread (Vries et al. 2015:105). Communities in CL-endemic areas must pay special care to decrease the risk of sandfly bites, especially at night. Because sandflies are so little, bed nets used to combat malaria can't keep them out unless the mesh is three times smaller. In addition to this it is vital to use insecticide treated bed nets so that maximum reduction of sandfly bite is accomplished (Vries et al. 2015:105).

2.3.7.3. Disease surveillance

Disease surveillance can help control epidemics and situations with high case fatality rates. Countries where CL is endemic can also benefit trough setting of efficient surveillance systems since information for constructing control measures can be available tought it (WHO, 2020).

2.3.7.4. Control of reservoirs

Animal reservoir host controls should be specifically designed for the area due to the complex nature of such schemes. It is also difficult to measure the efficacy of interventions based on the control of natural reservoir of Leishmania. Some studies reported the effectiveness of reservoir control to prevent leishmaniasis. In contrast, no evidence could be shown on the effectiveness of these methods in other studies. Overall, sensible proof on the success of interventions aimed at control of reservoir hosts is missing (Vries et al. 2015:105).

2.3.7.5. ***Social mobilization and strengthening partnerships***

Lack of vaccine and effective drugs for the highly prevalent parasitic diseases like CL forced the World Health Organization to relay on health education as the main preventive strategy. However, to bring favorable change in behaviour of the community, mobilisation and education interventions must be specifically designed the local community (WHO, 2020). Moreover, several research on vector control for leishmaniasis have stressed the need of health education and community participation in the disease's control. Training the community about preventive measures is very economical. Individulas in CL endemic areas can be alerted about the disease so that behaviors like use of preventive measures, and tseeking treatment can be improved (Nazari, Taravatmanesh, Kaveh, Soltani & Ghaem 2016:2).

2.3.7.6. Future preventive measures

Leishmania parasites are identified to cause little immune stimulation because they immunosuppressant and antigenic epitopes continuously vary. These immune-evasive attributes inflict a major difficulty on the progress of making a successful CL

vaccine. If a person recovers naturally, the immune system will produce defence mecanisms that lasts for life indicating the possibility of vaccine production. This has led to substantial investigation attempts in this field. Though the future is focused on vaccine development, for now vaccines for controlling human CL are not available (Vries et al. 2015:106).

Generally, to find effective and sustainable control and prevention strategy, cooperation between researchers, medical physicians, animal health experts, and public health agencies is essential (Alidadi & Oryan 2014:2).

2.4. CUTANEOUS LEISHMANAISIS IN ETHIOPIA

CL in Ethiopia is a zoonotic illness caused mostly by *Leishmania aethiopica*, with bush hyrax (Heterohyrax spp.) serving as the animal reservoir and known sandfly vectors *Phlebotomus longipes*, *P. pedifer*, and *P. sergenti* (Pareyn, Bosch, Girma et al. 2019:2). However, knowledge of environmental factors affecting transmission of CL in the country are poor. Therefore, successful control requires an integrated tactic with an improved knowledge of sociodemographic and environmental contributing factor (Seid et al. 2014:1).

2.4.1. Epidemiology of CL in Ethiopia

In 1913 an Italian epidemiologist Martogilo described CL in Ethiopia. But CL has been long known by many local names as "Kunchir" in Gojam, "Finchoftu" in central Shoa, Gonder, and parts of Wollo, "Chewie" in Sodo, "Shahegne" in north Shewa, "Volbo" in Ocholo, "Giziwa" in Tigray and "Simbirahalkani" in Wollega which is evidence for its long existence (Seife, Ayal, Misganaw & Zewdu 2018:1). CL is endemic to the country's highlands, which range in elevation from 1400 to 3175 meters above sea level as these areas comprise condusive environments for the survival of reservoir hosts. Ethiopia is among the high CL burden countries with 20,000 to 30,000 cases per year (Tamiru et al. 2019:1).

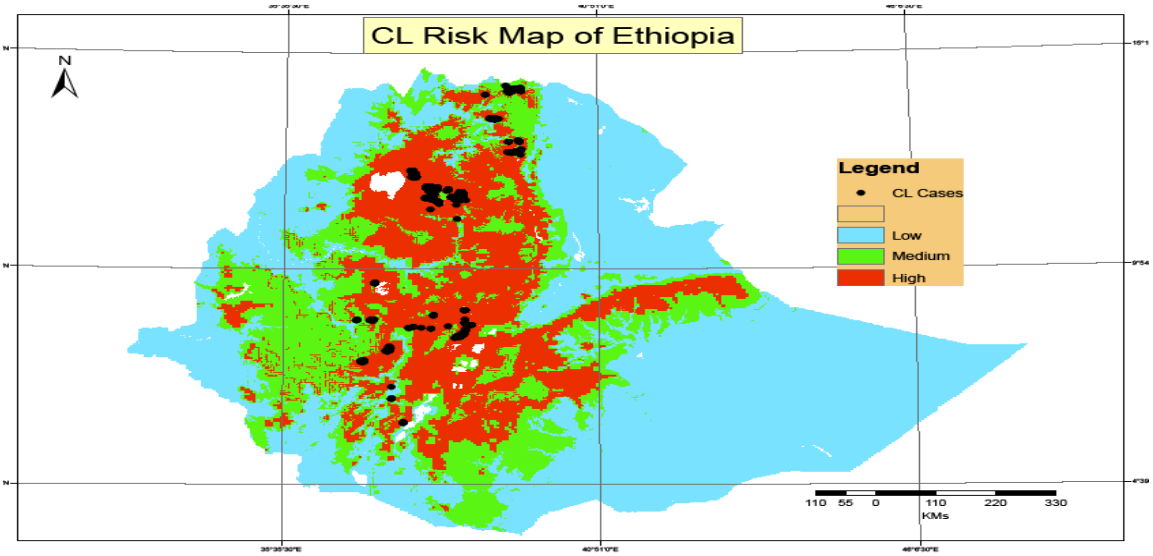


Figure 2.12: Risk map of CL in Ethiopia (Ethiopian Neglected Tropical Disease master plan: 22).

Seid et al. (2014) conducted study to identify the people and land area in the country at risk of CL, taking into account several environmental parameters. According to this study, over thirty million Ethiopians are at risk of contracting CL. These people are primarily from the highlands of Tigray, Amhara, the SNNP' Region, and Oromia. Slope, height, and yearly rainfall were found to be excellent predictors of CL presence in this investigation. Figure 2.12 depicts the risk of CL in Ethiopia.

In Ethiopia, substantial CL research has been carried out in the Rift Valley's western plateaus and lake basins. However, studies in key transmission areas such as Ochollo, Silti, and Sodo focus on the Rift Valley cliff above Lake Abaya, in the Kutaber zone in the eastern Ethiopian highlands near Dessie, in the Aleku zone of Wollega, the south-west plateaus of Bale and Sidamo, in Sebeta town near Addis Ababa, and in the eastern Tigray highlands of Adi-grat and Saesie Tsaed (Gebremichael 2018:433). The prevalence of CL and incidence rate in Ethiopia is increasing over the years as outbreaks are seen in places previously not known to be endemic (Negera et al. 2008). The incidence rates of CL in Ethiopia are presented in Figure 2.13 below.

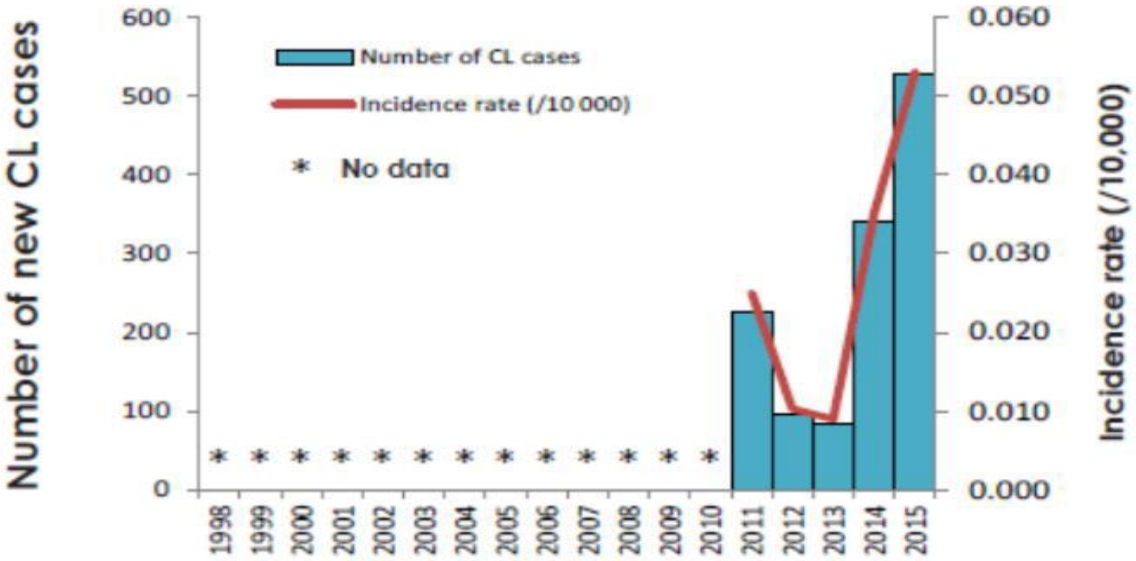


Figure 2.13: Incidence rates and number of new cases in Ethiopia (WHO, 2017).

CL is primarily produced by L. aethiopica (99.9%) in Ethiopia, however *L. tropica* and *L. major* are occasionally found in lowland areas. *Phlebotomus pedifer* is a species of Phlebotomus. In Ethiopia, the CL vectors *P. longipes* and *P. pedifer* are well-established (Bsrat, Berhe, Balkew, Yohannes, Teklu, Gadisa, Medhin & Abera 2015:2). *P. sergenti*, *P. duboscqi*, and *P. papatasi* have also been implicated as CL vectors in another investigation (Meles 2018:7).

Rock hyraxes have been identified as reservoir hosts for *L. aethiopica* in the CL endemic Ethiopian highlands. Severe CL outbreaks are frequently linked to the presence of hyraxes in highland locations, as hyraxes' ecological and behavioral characteristics make them ideal reservoir hosts for *L. aethiopica* (Gebremichael 2018:434). *L. aethiopica*-infested rock hyraxes were identified, demonstrating that the parasite has a zoonotic cycle. Two rock hyrax species, Procavia capensis and Heterohyrax brucei, are naturally hosts for *L. aethiopica* (Figure 2.14). Different types of hyraxes, however, are part of the CL epidemiology (Gebremichael 2018:434).

Figure 2.14: Reservoir hosts of CL in Ethiopia, above: Heterohyrax brucei (Shoshani 2005) and below: Procavia capensis (Linderman 2011).

Other hosts that can be considered as a reservoir animal in Ethiopia are bats. Two species of Leishmania namely *Leishmania major* and *Leishmania tropica* were identified from bats naturaly infected with the disease. This is a sign that bats could harbore Leishmania parasite and serve as disease reservoir in the country (Kassahun, Sadlova, Benda, et al. 2015).

2.4.2. Management of CL in Ethiopia

In 2006 Ethiopia produced the primary guideline which gives direction for specific diagnostic, management and prevention measures of VL. This guidline was then updated in 2013 with inclusion of CL. The currently used approach for the diagnosis of CL in the country is clinical assessment followed by laboratory verification via microscopic tests of samples from skin lesion. However, since 2014 only very few health centers in the country diagnose leishmaniasis (Yohannes et al. 2019: 2). Clinical symptoms are key in the diagnosis of CL even though verification is essential via parasite presentation from skin lesion. Localized Cutaneous Leishmaniasis (LCL), Mucocutaneous Leishmaniasis (MCL), and Diffuse Cutaneous Leishmaniasis (DCL) are the three clinical types of CL in Ethiopia (DCL). Furthermore, an uncommon form of the disease known as recidivans has been

discovered in Ethiopia (Dassoni et al. 2017). The importance of identifying clinical presentation in each of these CL forms is critical, and the second Guideline, published in 2013, specifies how to conduct clinical examinations of diverse CL forms in Ethiopia (Guideline for diagnosis, treatment & prevention of leishmaniasis in Ethiopia, 2013:33).

If a clinical examination cannot confirm the diagnosis, a parasitological test should be conducted. Aspiration, tissue biopsy, and skin scraping can all be used to acquire samples from questionable lesions. Samples should be obtained from the active area of the lesions, close to the lesion's margin, rather than from re-epithelialized lesions or scars. Slit skin smears with Giemsa's stain can be done at the health center level (Guideline for diagnosis, treatment & prevention of leishmaniasis in Ethiopia, 2013:33).

The main objectives of treatment in CL are to eliminate the parasite, control spread, mainly in the mucosal involvment, to speed up healing process, and to decrease scar formation scarring, particularly in cosmetic sites. In addition to curing patient proper treatment prevent disease re-occurence. According to the guidline mentioned above, there are three alternatives for the treatment of CL in Ethiopia. These are:

- To deny therapy;
- Application of treatments on the affected skin; and
- Use of systemic medicines.

Factors like lesion type and patient tendency to complete treatment affect choice of treatment options. However, available articles on the effects of this treatment option for CL in Ethiopia or any critical evaluation of the outcomes of any treatment method are scarce (Guideline for diagnosis, treatment & prevention of leishmaniasis in Ethiopia, 2013:35).

Directions put for prevention and control of CL or leishmaniasis in general in Ethiopia are vector control, awareness creation and community mobilisation via the

health development army and strengthening information system. Vector control can be targeted against the immature stages and adult sandflies. Unlike with malaria parasite, immature stages of sandfly are unreachable because of poor understanding of their breeding sites (Vivero, Torres-Gutierrez, Bejarano, et al. 2015:2). There are different strategies set for control against adult sandflies such as environmental management, insecticide application, Use of Long-Lasting Insecticide Treated Nets (LLITN), repellents (Chemical or Natural), and integrated control (Guideline for Diagnosis, Treatment & Prevention of Leishmaniasis in Ethiopia, 2013:39 &40).

Fundamental issue for any disease preventive and control plan is educating the community and propoting health. The health education program is effective if health care professionals and communities in prevalent areas are targeted. In Ethiopia, the introduction of the health extension programme is extremely valuable for disease control programmes which can help easily engage the community. Obviously, control interventions arranged at community level are feasible. In endemic districts of Ethiopia, teaching lessons for health agencies and health workers should be organised prior to the start of comprehensive control programme commences (Guideline for Diagnosis, Treatment & Prevention of Leishmaniasis in Ethiopia, 2013:40 &41).

In developing countries data collection and analysis is crucial for program implementation because of inadequate disease Information and poor surveillance system. Integrated Disease Surveillance (IDS) is the main approach and backbone of disease inspection in Ethiopia. Therefore, leishmaniasis being a disease with frequent eruptions, must be incorporated in the IDS system of regions with widespread CL occrance (Guideline for Diagnosis, Treatment & Prevention of Leishmaniasis in Ethiopia, 2013:40 &41).

2.4.3. Prevalence and species causing cutaneous leishmaniasis in Ethiopia

CL is known to be endemic in Ethiopia in magnitudes of undetermined prevalence and distribution (Dawit & Shishay 2014:96). The excessive price of treatment and

time wasted for the period of hospitalisation are the major economic impact of the disease in Ethiopia. The illness affects the rural poverty-stricken population and normally, an outbreak follows harvesting periods (Dawit & Shishay 2014:96).

The most recent outbreak of CL was reported in the Silti District of the country's south, which was previously not recognized as being CL endemic, with a prevalence of 4.8% and the causative agent identified as *Leishmania aethiopica* (Negera et al. 2008). A study was conducted in Tigray region and the overall prevalence is 14.0% where 6.7% and 7.3% carry active lesion and scar, respectively. This study also pointout that with the highest prevalence is amongst 10–19 years age groups and Leishmania species identified was *L. aethiopica* (Bsrat et al. 2015).

CL is seen in 5.6% of patients at Ayder referral hospital in Mekelle, Tigray, Ethiopia (Tilahun et al. 2014). Only 11 (0.6%) of 1869 children tested positive for CL in a study to examine patterns of skin infections in children visiting a dermatology clinic in Wolaita Sodo, southern Ethiopia (Kelbore, Owiti, Reid, et al. 2019:4). The total prevalence of CL was 65.8% among primary students in Ochollo town Southwestern Ethiopia. Of these, 59.9% were observed to have an active lesion while 4.01% present a scar (Bugssa, Hailu & Demtsu 2014:113).

Investigation of CL in Northcentral Ethiopia reported a prevalence of 1.5% with an increasing trend from 2012 to 2018 (Eshetu & Mamo 2020). The prevalence of CL was 22.4% (46/205) amongst clinically presumed patients visiing Borumeda Hospital in Northeast Ethiopia (Bisetegn, Zeleke, Gadisa, et al. 2020).

2.4.4. Risk factors for transmission of cutaneous leishmaniasis in Ethiopia

Many risk factors, including environmental conditions, human behavior, economic situations, immunogenic status, and genetic concerns, have contributed to the growth in leishmaniasis cases. House structures associated with low socioeconomic class, such as those with broken mud or thatched-coated walls, and moist mud

flooring, could pose a CL danger. Sandfly vectors hide in fissures and crevices in the unprotected house walls, ceiling, or floor. Sandfly presence and vector profusion can be accelerated and enhanced by behavioral and work-related behaviors such as sleeping on the floor or outside, and plants adjacent to the house (Oryan, Alidadi & Akbari 2014:1). Figure 2.15 shows different factors playing a role in the transmission of CL.

Furthermore, residing near to a person with previous case of leishmaniasis and lack of insecticide spraying in the houses strongly increases infection risk. Sleeping conditions such as sleeping outside without bed nets can increase the risk of sandfly exposure (Oryan et al. 2014:1). Several studies in Ethiopia have identified factors linked to the frequency of CL in various populations. Environmental factors such as the house's proximity to a canyon where hyraxes live, the presence of Sensel (Adhatoda schimperiana) and Acacia trees, and the presence of domestic animals with people were all linked to the development of CL (Negera et al. 2008). Environmental factors like the presence of animal burrow, cave or gorge near residents, reservoir hyraxes, domestic animal dung and farmland near residents' houses were indicated to have a significant association with CL (Bsrat et al. 2015). Elevation, slope, and yearly rainfall have also been found to be good predictors of CL presence (Seid et al. 2014:377). Being a farmer, gender, location of the residence adjacent to both farm and gorge, and the presence of the hyrax all had statistical relevance with the prevalence of CL in the northern portion of the country (Tilahun et al. 2014).

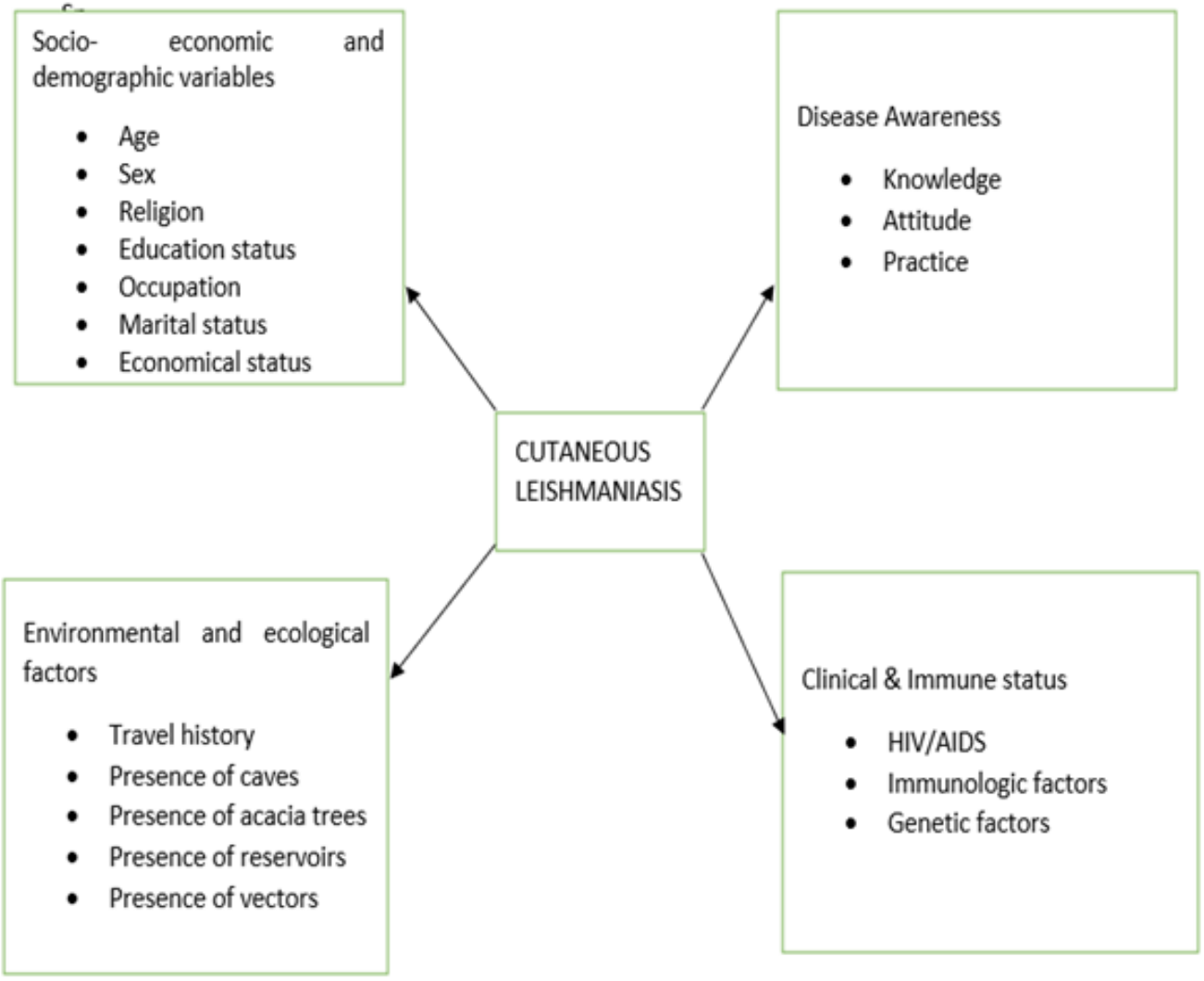


Figure 2.15: Factors affecting the occurrence of CL (Modified from Tilahun et al. 2014:192).

Poor housing conditions, migration for employment, desertification, immunosuppressive diseases such as HIV and AIDS, and malnutrition are some of the risk factors connected to the prevalence of leishmaniasis in Northwest Ethiopia (Tamiru et al. 2019:9). In addition, having no bed net, having irrigation nearby, and traveling to CL endemic zones are all linked to CL (Eshetu & Mamo 2020). Working in and near agriculture, having a residence next to a farm, the presence of a gorge in the village, and the presence of hyrax in the village are all statistically linked to CL prevalence in Northeast Ethiopia (Bisetegn et al. 2020:6).

2.4.5. Knowledge attitude and practice of cutaneous leishmaniasis in Ethiopia

The evaluation of baseline familiarity of the endemic residents is the first stage in the control of CL. This is followed by an examination of socio-demographic and environmental risk variables in order to build integrated preventative and control techniques. In each endemic area transmission cycle of CL is different which makes comparative use of data from one area to another problematical (Nandha, Srinivasan & Jambulingam 2014:1055). Therefore, the KAP survey in each endemic area will be vital to design intervention strategies against the disease. Different studies conducted in this respect reported various findings.

In an Iranian study of CL-infected children's moms, it was discovered that 40% of them had a poor comprehension of the disease. When it came to topics like the likelihood of reinfection, potential reservoir, time of spread, sandfly features, sandfly behavior, the difference between rural and urban leishmaniasis, vector control methods, and, finally, the use of sandfly monitoring methods like insecticide-treated bed nets, there was a significant lack of data (Hejazi, Hazavei, Bidabadi, et al. 2010). According to the survey, over 28.9% of moms had incorrect KAP scores about leishmaniasis, implying that one out of every three Isfahan residents requires practical education on leishmaniasis prevention (Hejazi, Hazavei, Bidabadi, et al. 2010).

A similar study conducted in India presented that 39.8% of participants recognised images of CL shown to them nonetheless these did not have good beliefs. In addition, these populations have poor understanding of vectors, disease spread, risk factors and control measures against CL. Lack of knowledge on vector and cause of CL was seen among this communities which could lead to increased risk of CL dues to reluctant use of personal protective measures (Nandha et al. 2014). According to the findings of a survey conducted in Iran, the participants' knowledge was below average (Rakhshani, Kashfi, Ebrahimi, et al. 2017). Unsatisfactory practices regarding CL were reported by different authors. A study conducted in India indicates that utmost 2% of participants use mosquito nets (Nandha et al.

2014:1054). In Ecuador practices for treating lesions such as the use of acids, lighted matches and gasoline are possibly dangerous (Nandha et al. 2014:1055).

Knowledge of CL and sandfly association were linked to the use of sandfly management strategies in Columbia (Pardo, Carvajal, Ferro, & Davies 2006) and Guatemala (Arana, Rizzo, Navin, Klein & Kroeger 2000). A comparable research of CL patients' KAP found that the majority of them referred to the condition as a "one-year sore," linked it to insect bites, and believed it was not contagious (Abazid, Jones & Davies 2012). The majority of participants believed it could be avoided by using bed nets and insecticides, and they were also aware of the treatment options (Abazid, Jones & Davies 2012). An investigation of the knowledge, attitude, and behavior of health care providers in tertiary health care services in Nigeria revealed increased detection of CL lesions as well as precise awareness of its spread and prevention. This could be obvious as the study population are health professionals. However, inadequate understanding and practice of CL treatment are observed among these (Awosan, Isah, Alayande, Enokela, Makusidi, Agwu, Abubakar, Abdullahi & Aderahman 2013).

In the Knowledge attitude and practice study in Northern Ethiopia, most respondents can identify the CL. In contradiction, these communities were also shown to have poor knowledge of CL cause and transmission as they wrongly perceive CL to be a genetic related illness. With views to medication, 99% of CL suspected patients use a traditional medicine such as the usage of herbs, and holly water called “Tsebel” in Amharic. Totally all of them do not know about modern medical treatment (Bsrat et al. 2015:6).

In a KAP study done in southern Ethiopia, 67.6% of the respondents heard about CL and the rest are anawere of it. More than half (54.8%) of the study members who have heard about CL had a desired attitude that CL was a challenge in their locality but undesired attitude for medication of CL. Roughly, half of these answered that CL is avoidable. Most of the respondents do not practice outdoors sleeping and

do not also sleep nearby vegetation with or without a bed net (Kebede, Worku, Ali, et al. 2016).

In a recently conducted KAP survey in CL endemic area, Northwest Ethiopia, 21.6% confessed that they have learned about sandflies however nearly (46.3%) of them are not awere of the peak transmission time (Tamiru et al. 2019). Major clinical presentation of CL for 47.1% of the respondents was scarring lesions and 51.2% think that CL can be best treated using traditional medicines. Barriers for use of modern medication are accessibility, distance of treatment facilties and long duration of modern treatment (Tamiru et al. 2019).

2.5. CONCLUSION OF LITERATURE REVIEW

This chapter provided details of literature about leishmaniasis in general and CL in particular. Different clinical presentations of the disease were discussed. Vectors and reservoir hosts of the disease were reviewed along with their distribution throughout the world. The burden, management approaches, contributing risk factors and epidemiological studies of CL in Ethiopia were presented. The following chapter will illustrate the investigation approach.

CHAPTER 3

RESEARCH DESIGN AND METHODS

3.1. INTRODUCTION

In Chapter 2, the results of approximately 100 literature reviews were organized topically. A cross-sectional descriptive approach was utilized as the primary quantitative tool in this investigation. Detailes of the research approach will be descriped in the resecrh design section 3.2. of this chapter. The study was conducted in different phases using different approaches such Questioner administration, household survey and laboratory analysis. The questionnaire administration was used for knowledge attitude and practice survey and identification of different risk factors. Household survey was used for identification of individuals with CL case (active and scare lesions) to investigate the disease prevalence, collection of tissue samples and laboratory diagnosis to identify the Leishmania parasite species.

As defined in Chapter 1, the study objectives were

Phase 1: -

- To access knowledge, attitude, and practices of Sodo community on CL.

Phase 2: -

- Investigate demographic, household and host-related risks related to transmission of CL.
- To clarify the role of animal and environmental factors on spread of CL in the district.

Phase 3: -

- To determine the prevalence of CL in the area.
- To identify the Leishmania species circulating in the community.

This chapter covered the research design, study population, sample size calculation, data collecting, sampling technique, and data analysis in order to achieve the above-mentioned objectives. The methods for addressing ethical concerns are also demonstrated. The study's internal and external validity are also discussed.

3.2. RESEARCH DESIGN

Research design is a constructed plan of activities followed to resolve research questions. Planning a condusive research design includes setting ahead the type of data to be obtained, identifying the study area, timetable of the study, source of participants, the dependant and independent variables, method of data collection, the theorthical hypothesis and data management and analysis methods (McCombes 2019). In research design, project parameters to be included in the study and standard for evaluating results and drawing clonclusions will be drafted. The way of data collection, measurment, analysis, and interpretation are important for considering the research valid and reliable. Successful research proposals, scientific papers, or dissertations need to have a strong research design (McCombes 2019). In quantitative studies, it is advised that overall research approaches be setted priorly focusing on collecting and analysing of numerical data. The next step in the research design is planing of participant selection, setting data collection methods, identifying data analysis approach and composing the report on the results (Creswell & Creswell 2018: 43)

Established on this respect, this research utilised a quantitative study approach to answering the research questions. The focus of quantitative methods is measuring objectives and examination of numerical data obtained by adminstering questioners or surveys unlike qualitative and quantitative research focuses on the gathering of numerical figures which will be used to draw conclusions about a group of individuals or explain a particular phenomenon. The two types of quantitative study approaches are descriptive where individuls or events are evaluated once to establishes associations between variables and in experimental study subjects are assessed prior to and following intervention for identifying causality (Sheard 2018).

There are two types of quantitative research: cross-sectional and longitudinal. Cross-sectional studies collect data at a single point in time, whereas longitudinal studies collect data multiple times from a small group of people who share common qualities. Cross-sectional study designs are often used in

public health planning to assess the frequency of the outcome of interest in each group of people (Levin 2006:24).

In this study, primary quantitative method using a cross-sectional descriptive approach is applied.

3.3. RESEARCH METHOD

3.3.1. Study area

The study was conducted in the Sodo District, which is located in Ethiopia's southern area (Figure 3.1). Regional states are Ethiopia's primary administrative divisions, followed by Zones, Weredas (Districts), and Kebele. As a result, Kebele is the country's smallest administrative entity. There are nine regional states based on ethnic territoriality (called Kililoch in Amharic). Gambela, Afar, Amhara, Somalia, Oromia, Benishangul-Gumuz, Harari, Southern Nations Nationalities and Peoples' Region, and Tigray are the countries in question. Aside from that, the two chartered cities are Addis Ababa and Dire Dawa (Central Statistics Agency 2012).

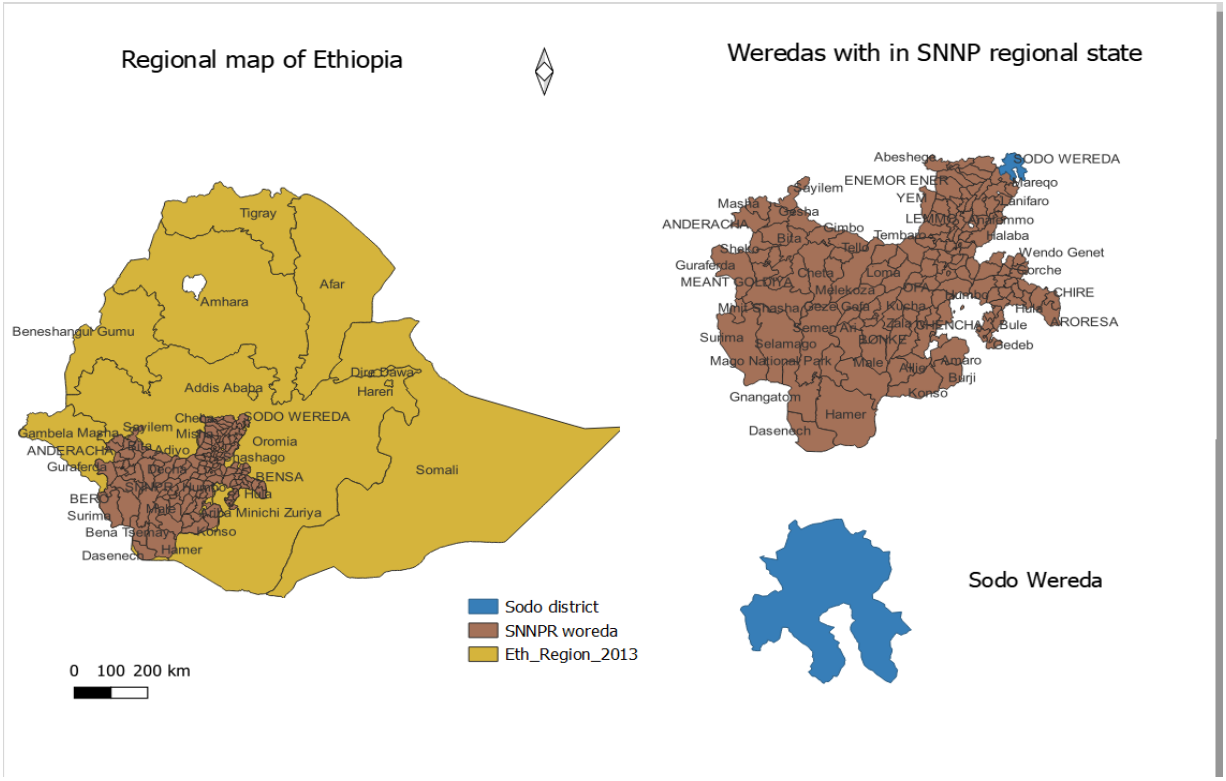


Figure 3.1: Map of study area.

The research was conducted in the Sodo District. Sodo is a district of Ethiopia's SNNP Regional State's Gurage Zone. Meskan district borders the district on the south, and Oromia region on the west, north, and east. Buee, the administrative capital of Sodo, has 109,943 inhabitants and spans an area of 109,943 hectares. The area is around 178 kilometers from Awassa, the region's city, and 105 kilometers from Addis Ababa, Ethiopia's capital. The district is expected to have a total population of 175,725 people, with 89,619 (51%) women (2007 National Census estimated for 2012/2013) and 90% of the population residing in rural regions (2007 National Census predicted for 2012/2013) (Central Statistics Agency 2012).

Almost everyone in the Sodo population (93.0%) practices orthodox Christianity. The Sodo Gurage people are the majority, followed by the Oromo and Amhara peoples. There are four urban kebeles and more than fifty rural kebeles in the district (Kebele is the last administrative components in the government structure). Both highland and lowland kebeles make up the rural section of the area. The district is scheduled to have eight health centers, 55 health posts, two private clinics, and three pharmacies (Boru, Shimels & Bilal 2017: 528).

Based on traditional healers, district health professionals, and secondary data six kebeles, namely, Kela Zuria, Kola Nurena, Michael Semero, Adazer, Beka, and Genete Mariam were selected with a total household of 2632. The number of households in each kebele is depicted in Table 3.1.

Table 3.1: Total number of households and population in selected kebeles.

Keble	No of households	Population
Kela Zuria	333HH	2028
Kola Nurena	362HH	2389
Michael Semero	475HH	3055
Adazer	610HH	3477
Beka	299HH	1678
Genete Mariam	553HH	2912

3.3.2. Source population and study population

The source population is the group from which study participants are drawn. The study's source population was all residents of the six kebeles chosen (Kela Zuria, Kola Nurena, Michael Semero, Adazer, Beka, and Genete Mariam). In descriptive studies, the study population needs to be defined so that samples will be taken from it. Variables such as age, location, sex, occupation, ethnic group, and religion can be used to define a study population (Banerjee & Chaudhury 2010). As a result, the study population included all households chosen at random as well as those living in those households.

3.3.3. Sampling and sampling frames

The sampling frame includes all households in the district.

3.3.3.1. Sampling for knowledge attitude and practice questionnaire survey

A knowledge, attitude, and practices (KAP) survey are used to gather data on what is known and believed by people about a specific topic. A KAP study is a quantitative method because it uses standardised questionnaires to reveal misconceptions or misunderstandings about factors affecting implementation activities or potential barriers to behavior change (Alhaj 2018).

Alhaji (2018) indicated that a KAP survey can be used to:

- Quantify known situation, test a hypothesis; generate new point of view about a situation.
- Understand and modify knowledge, attitude, and practices health-related or other subjects.
- Generate baseline data and evaluate the effectiveness of health education interventions to change health-related behaviours.
- Design locally applicable intervention strategy and plan activities that are suited to the respective people involved.

For determining the sample size for the KAP questionnaire study, 50% of the communities are assumed to be knowledgeable about CL with 2% marginal error

and 95% confidence interval. The following single population proportion formula was employed to get the definite sample size with a 10% non-response rate.

$$n = \frac{(Z\ 1/2)^2 \times (pq)}{d2}$$

Where: d= Degree of marginal error (5%)

Z = level of confidence (95%)

P = CL prevalence (50%), q = 1-p

adding 10% non-response rate,

$$n = \frac{1.96^2 * 0.5(1-0.5)}{(0.5)^2}$$

The required sample size was 423 households.

3.3.3.2. Sampling for risk factors and prevalence survey

Since the KAP survey and the risk factor identification survey were administered in different years – 2017 and 2018 respectively, it was decided to select another set of study population for the six kebeles for identification of factors for transmission of CL and estimation of the prevalence of the disease in the study area. Accordingly, sample sizes were defined on the basis of previously set prevalence rate of 65.8% from Ochollo, southern Ethiopia (Bugssa et al. 2014:1) with a degree of significance α 0.05 needed precision and adding 10% non-respondent rate, the formula

n = Z (α/2)2P (1-P)/ d2, Where:

n = Sample size

d = level of precision

Z (α/2) = limit of confident

P = previously set prevalence rate of CL

The computed sample size holding d=0.05, is:

Hence $n = \frac{(1.96)^2 \times (0.66)(1-0.66)}{(0.05)^2}$

Giving a overall sample size of 379

3.3.3.3. Sampling for species Leishmania identification

While administering questionnaires to identify disease transmission risk characteristics, samples were taken from clinically questionable subjects for species identification. The sample size was calculated using the single population proportion approach, assuming that 95% of the dominant species in the research region fit to L. aethiopica (Bsrat et al. 2015:2) with a 5% level of precision and a 95% confidence range.

$$n = \frac{(Z\ 1/2)\ 2\ (pq)}{d^2}$$

using the above formula, the sample size will be 73. Though this is the required sample size for identification of Leishmania parasite, clinical samples were collected from 49 individuals because only this number was found with active CL during the survey.

3.3.4. Sampling procedures

To sample 423 HH for the KAP survey and 379HH for risk assessment and prevalence study, simple random sampling was employed after proportionally allocating the samples to be taken according to HH size in each kebeles then in each Gote (Subdivisions of Keble). The general presentation of the sampling procedure is presented in Figure 3.2.

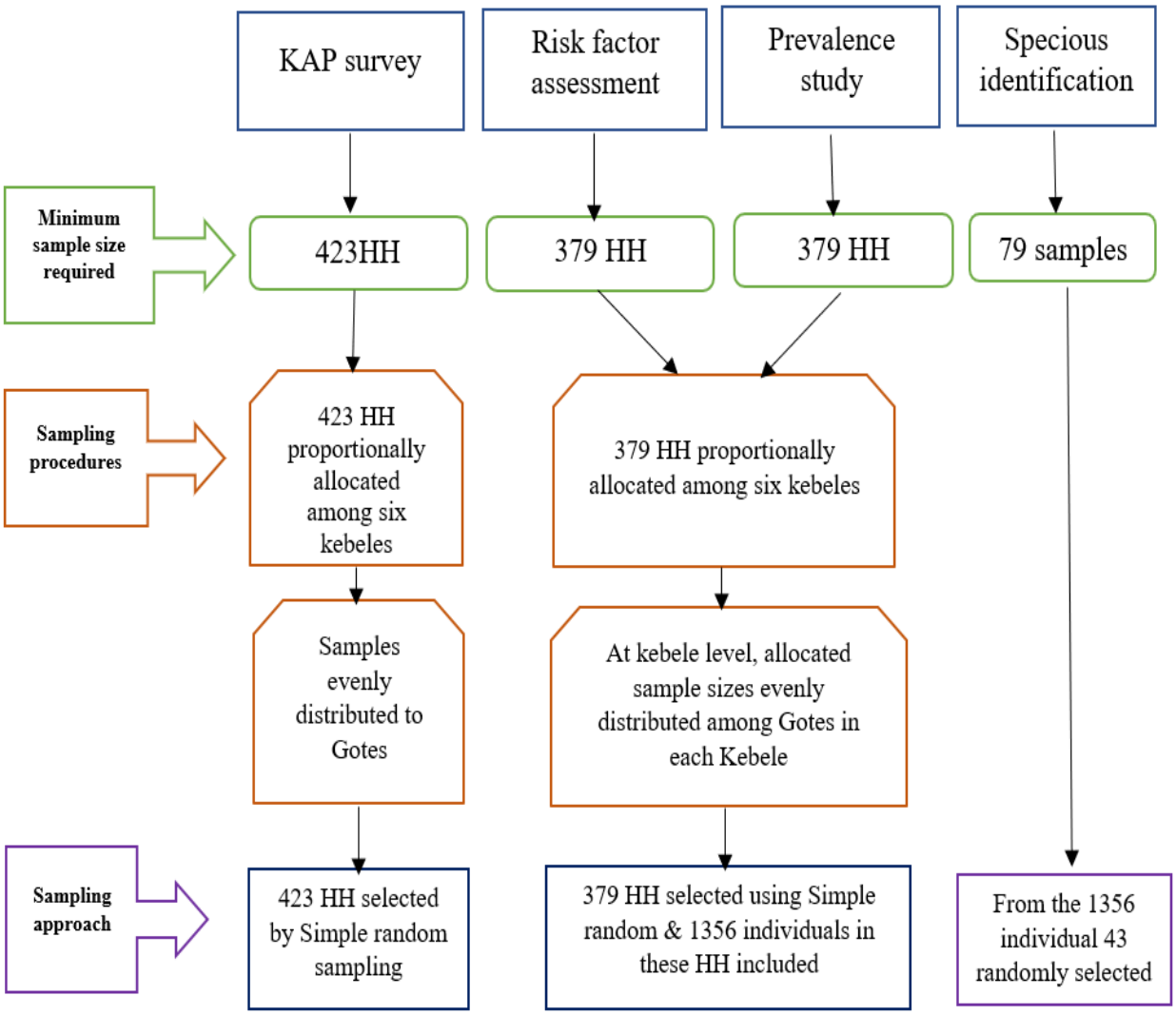


Figure 3.2: General presentation of sampling procedure and approach.

3.3.4.1. Sampling procedure for the knowledge attitude and practice survey

For the KAP questionnaire survey dividing the total sample (423HH) to each kebele was done by applying the following population proportion formula

$$n_1 = \frac{\text{Number of HH in the kebele X Desired sample size}}{\text{Total number of HH in the 6 kebeles}}$$

For instance, Kela Zoria= 333/2632x423=54 samples to be taken from this kebele and this was further subdivided by the 8 Gotes (Gote a name given to locally set subdivision under the smallest administrative unit, kebele) in the kebele which finally comes to six questionnaires to be administered to each Gote. And for the first Gote in Kela Zuria kebele for example Duleti1 which has 40HH we will take (40/8=5. So, every fifth household will be visited and if the fifth household is not available the immediate next house will be considered). The number of households in the remaining five kebeles is 360 (Kola Nurena), 470(Semero), 607(Adazer), 296(Beka), and 552(Genete Mariam).

Similar calculations were done to the other five kebeles

- Kola Nurena 58 samples (with 9 Gote and 6.4 questionnaires /each Gote)
- Semero 76 samples (with 9 Gote and 8.4 questionnaires /each Gote)
- Adazer 98 samples (with 8 Gote and 12.25 questionnaires /each Gote)
- Beka 48 samples (with 8 Gote and 6 questionnaires /each Gote)
- Genete mariam 89 samples (with 9 Gote and 9.8 questionnaires /each Gote)

3.3.4.2. Sampling procedure for risk factor assessment and prevalence study

In Sodo woreda, the population of six kebeles was examined. A total of 379 households and 1,356 people (all from the same household) agreed to take part in the research. Individuals with a probable case of leishmaniasis were included, and clinical assessments were performed. The 379HH were chosen at random using the population proportion formula below, as used in the KAP survey.

$$n_1 = \frac{\text{Number of HH in the kebele X Desired sample size}}{\text{Total number of HH in the 6 kebeles}}$$

All individuals (1,356) who consented to join in the study from the 379HH were contained and the proportion of these individuals in the six kebeles is presented in Table 3.2.

Table 3.2: Distribution of sampled individuals among selected kebeles (N=1356).

Residence Kebeles	No individuals in households (%)
Adazer	509(37.54)
Beki	71(5.24)
Genete Mariyam	326(24.04)
Kela Zuria	171(12.61)
Kola Nurena	98(7.23)
Michael Semero	181(13.35)

For the prevalence study, 123 CL cases with the clinical presentation of the disease 49 skin scrapping sample from persons showing active lesion of CL were selected for species identification.

3.3.5. Inclusion and exclusion criteria

For the KAP survey and risk assessment study, all individuals of a selected household were eligible. Participants comprised any consenting individuals residing in the selected six kebeles. Individuals who live outside the defined areas were excluded to avoid sampling of non representative individuals. Households that have been visited three times without finding an adult to approach for consent were considered non-responsive, excluded from the study, and replaced with an alternative household.

Only people with suspected CL were considered for sample collection (skin scrapings) for species identification purposes, with other dermatological disorders (such as skin TB and scabies) being rejected by certified health professionals based on clinical circumstances. This is done to prevent non-CL samples from being collected. Only individuals who gave informed consent/assent had their samples taken (Annexure J).

Because of the possibility of culture contamination, study participants whose lesion was contaminated by secondary infection were not allowed to give skin scrapings. Patients were excluded from the study if they were found to have a secondary

bacterial or fungal infection or were using active CL therapy, as treatment may influence lesion and parasite load.

3.3.6. Data collection

3.3.6.1. Preliminary survey

A panel of experts, including the principal investigator and senior scientists (advisors), visited to the zone before deciding on the study sites and kebeles. The Zonal Health Bureau was consulted in order to have a broad understanding of the disease and to make the study more efficient. Six kebeles were chosen based on the district's health profiles and discussions with the district's traditional healers. Each kebele's health center and the Sodo District Health Office were called. Even though the Health Office was aware of the disease's prevalence, there was no documented case history.

3.3.6.2. Knowledge attitude and practice questionnaire administration

The KAP questionnaire (Annexure A & B) was pre-tested to maximise the quality of data by finding out how well the questions are understood by the interviewees. Pilot testing is a technique used to test accuracy of study instruments such as questioners so that it could be refined by fixing vegue and erranous parts to effective implementation of the main study (Lobiondo-Wood & Haber 2002:199). Pilot testing is a tool that is used to to measure the application, suitability, competence, completeness, and indicative or personal hints. Furthermore, pretesting also seeks to see if questionnaires are clear, instructions are effective, responses are complete, time required to complete the questioner, reduce biasness and also correctness of questionnaire formats (Lobiondo-Wood & Haber 2002:199). More importantly pretesting is essential to spot unpredicted difficulties and errors, regulate the viability of the sampling method, recognise gross insufficiencies earlier execution of to full-scale study (Lobiondo-Wood & Haber 2002:199). The KAP questionnaire was pretested to confirm that evidence about all recognised subjects would be gathered completely. This was done to see if the questionnaires were clear comprising correct words which can be easilty and approoriately understood by the participants. All individuals who partaken in pretesting of the survey tool were

omitted from joining in the main research. For this purpose, 30 individuals were interviewed at the stage of pre-testing. The questionnaires were re-corrected and administered to the participants based on the results of the pilot testing.

Face-to-face interviews with household heads were conducted from January to April 2017. Environmental data was collected by analyzing the presence and absence of each variable for the household head. Demographic and socioeconomic data were collected directly from the household head. The questionnaire, which had well-organised questions, was used to collect data on socio-demographic characteristics, CL knowledge, a scale of attitude toward CL, and CL preventative practices in the English language. The questionnaire was further translated to local language (Amharic) before administration. Knowledge was measured using an 8-item questionnaire containing:

- If they could name, the disease after being shown a picture of CL manifestation
- Have you ever heard of CL?
- What is the cause of CL?
- What are major symptoms of CL?
- How is the disease transmitted?
- Whether they know the importance of the vector as a transmitter of leishmaniasis after being shown a picture of phletomine sandfly believed to exist in area
- Where do vectors breed?
- When does the vector prefer to bite?

The attitude was evaluated by using a 6-point questionnaire comprising:

- Do you think chewie can be treated?
- What is your preferred choice of medication for the treatment of Chewie?
- What is your opinion on the outcome of CL if left untreated?
- Do you feel that you are well informed about chewie?
- Willing to participate in CL control activities?
- Do you consider CL a serious health problem in your localiities?

Prevention practice had 8 items including

- How was Chewie treated?
- Use of Preventable action for CL?
- Type of methods used to prevent CL.
- Having bed nets in the house.
- Sleeping condition.
- Using bed nets when sleeping.
- Using repellents; and
- Working time preference at peak temperature.

The survey tool was translated to Amharic language keeping its original meaning. The household heads in the identified houses or another older family member, if the household head is not around, were interviewed. Data was collected by nine health extension workers, with a diploma and bachelor's degrees, were selected from each kebele of the Sodo District for this purpose. The data collectors received a one-day training which took one full day before the beginning of data gathering. The training which was offered by the principal investigator focus on about aim and importance of the study, how thay should approach respondents and how to inform and obtain consent from the respondents. During the actual study a one-on- one interviews were conducted after locating a suitable place in around respondent's residence. The local name for CL,"Chewi", was recognised during the pretesting and therefore this name was used to refer to the disease for the respondents.

Knowledge scores range from 0-8 and classified as unsatisfactory knowledge if the score is < 4 and coded as 0 while scores > 4 were set to be satisfactory knowledge with value code of 1. Attitude towards CL was measured by six questions therefore, scores between 0 and 3 were put as unfavorable attitudes but attitude score > 3 categorised as having favorable attitude. Practice against CL was assessed by eight questions and practice score of 4 and above were stared as good prevention practices.

3.3.6.3. Assessment of cutaneous leishmaniasis risk factors

House-to-house survey was done to investigate risk factors important in the transmission of the area from May to August 2018. A structured questionnaire (Annexure C & D) was prepared for this purpose and pretested following methods used in the KAP survey. These survey data were obtained by the same health extension workers mentioned in the KAP survey section. The questionnaire was divided into different sections that

- Access household factors (including wall type, wall condition, roof type, floor type, presence of functional latrine, and presence of screen on window/door);
- Environmental factors (the type of tree around the house, presence of farm, backyard and public assembly area around the house, the existence of cave or gorge and type of water source);
- Host factors (such as keeping dry wood inside the house, history of travel, sleeping condition, spending time near/at gorge early morning, irrigation of crops at night and use of personal protective measures); and
- Animal factors (presence of hyraxes, presence of domestic animals, the habit of dumping animal dung near house and presence of hyrax burrow).

Questions related to household factors were filled in by observation of the house condition and the surrounding. Questions regarding environmental factors and animal factors were delivered household head in each house because these factors are the same for all individuals in the household. However, questions about host factors were obtained from each adult individual participant and in the case of minors (under 18 years old) participants, the parent (mother and father) or guardians were interviewed.

3.3.6.4. Estimating of disease prevalence

Samples were obtained from clinically suspected CL participants in the family while visiting randomly selected households for a risk assessment study. On each person interviewed, a health care provider performed a physical examination to see if there were any lesions or markings indicative of scar formation. Individuals suspected of

having CL were invited to come to the Bue health center and offer a swab for laboratory testing. All individuals, whose samples were positive for *Leishmania,* were referred to ALERT (All Africa Leprosy Rehabilitation and Training Center) hospital for treatment. ALERT, which is in Addis Ababa, is one of the major hospitals for the treatment of CL in the country. The hospital currently has 240 beds for dermatology, ophthalmology, and surgery department. Patients with negative leishmaniasis tests were referred to surrounding (district or neighboring district) hospitals for proper diagnosis and treatment after consulting with the Bue Health Centre.

The questionnaire for patients suspected of being infected with CL was filled out using clinical examination for the presence and absence of possible leishmaniasis scars and lesions. For CL patients, a different questionnaire was created and filled out by interviewing them and doing a clinical assessment at the Bue Health Centre. Parents or guardians were engaged in the case of minors. A patient data sheet (Annexure E) was created to record diagnostic, clinical, demographic, and epidemiological data for the clinical profile CL. With an electronic input interface with Epi-Info version 7 and SPSS version 24.0, the surveys were designed to be simple.

3.3.6.5. Clinical sample collection

For parasitological identification, clinical samples of skin scrapings were taken. Clinically suspicious patients were contacted about the study before samples were taken, and only those who agreed to participate and grant permission were included. A pre-tested patient record form was also utilised to collect certain basic data, such as educational background, occupation, travel history, leishmaniasis treatment, and length of stay in the study area. A trained laboratory worker from ALERT hospital took the samples from the consenting suspicious research volunteers. The active portion of the lesion was identified and treated effectively with 70% ethanol before being allowed to dry. After cleaning the lesion portion with 70% ethanol socked sterile gauze, skin scraping samples were taken with a sterile disposable blade.

Inflamed parts of the lesion at the periphery of the lesion were preferred for sampling and a small skin slit scrap specimen was taken from the infected parts. If the person is presented with one or more skin lesions which appear more than two weeks ago on bare body parts such as arem, face, neck, and leg, then the person is considered to fulfil operational definition for CL (Aberra, Abera, Belay, Kebede, Gadisa & Tasew 2019:2).

Skin scraping samples were immediately injected in two copies onto Novy-MacNeal-Nicolle (NNN) medium in the field. On clean microscopic slides, a skin slit smear was also done in two copies. The NNN culture medium was placed in an icebox right away. Slides that had been smeared were placed in a slide box. The samples were then sent to the Armauer Hansen Research Institute (AHRI) in Addis Ababa's Leishmania laboratory.

3.3.7. Laboratory diagnosis

Smear and culturing samples from skin scrapings from Novy MacNeal Nicolle were employed in the laboratory for parasitological confirmation (NNN). For molecular identification of Leishmania species, PCR amplification and RFLP analysis of the ribosomal DNA Internal Transcribed Spacer (ITS) sequences were used. The Armauer Hansen Research Institute (AHRI) Leishmania Laboratory completed all laboratory work in August and September 2018.

3.3.7.1. Smear test

Giemsa stain is a thin and thick smear staining technique used for detecting blood parasites such as malaria, Leishmanaisis and other blood parasites. It can also be used to morphologically distinguish the nuclear and cytoplasm of red blood cells, white blood cells, Platelets, and parasites.

Skin tissues were sampled with a disposable lancet for the creation of stained smears. The tip of the blade was used to make a small incision on the cleansed edge of the nodules and lesions. Then, by twisting the blade 90 degrees, the lesion was scraped with the cut side of the incision to prevent and pick up the skin tissue.

The patients were then given a little amount of lesion material to spread on a glass slide. Furthermore, the smears created in the preceding method were fixed in methanol, dried in the air, and stained with Giemsa. The slides were examined under a binocular microscope at a magnification of 100x in oil immersion with the goal of finding Leishmania amastigotes (Mokobi 2020). The Leishmania organisms which are mostly positive with Giemsa stain, appear as a blue stain on paraffin-embedded sections.

3.3.7.2. Culture media preparation

Leishmania is isolated by a commonly and worldwidely known medium called NNN (Novy-MCNeal-Nicolle). This medium is particularly suitable for the growth of Leishmania organisms and gives rise to rise to a motile, extracellular form called, promastigote. McNeal and Novy developed the medium and Nicolle modified it later. NNN medium contains two phases, blood agar base (part A) and Locke's liquid solution, an overlay medium (part B). Since this blood meadia is nutritious it suitable for growing Leishmania and Trypanosoma. The amastigotes change to promastigotes after specimens are put in the liquid phase of the culture and incubated for 24 hours (Tankeshwar 2016).

Novy-MacNeal-Nicolle (NNN) medium (Difco, Franklin Lakes, NJ, USA) with Locke's solution (Sigma, St Louis, MO, USA) as an overlay with penicillin-streptomycin (100 IU/ml— 100 g/ml) and L-glutamine (0.29 mg/ml) (GIBCO, Paisley, UK) were used to culture clinical isolates and the reference strains of Leishmania (Bugssa et al 2014; Bsrat et al. 2015). The NNN medium was made by weighing, mixing, and dissolving the medium's constituents (9.2g nutrient agar (Difco), 0.6g D-glucose (anhydrous), and 2.4g sodium chloride (sigma) in 400ml distilled water in a microwave oven with continuous shaking every five minutes until a pure solution was detected. It was then autoclaved for 30 minutes at 121°C. The Ethiopian Public Health Institute provided aseptically collected sheep blood in a vial containing glass beads, which was shaken to defibrinate. On the day of collection, the blood was delivered to AHRI. The defibrinated blood was heat-activated in a 37°C water bath for one hour before being moved to a 56°C water bath for 20 minutes to inactivate the

complement at AHRI's Leishmania laboratory. At 500 degrees Celsius, the autoclaved components (400 mL) and heat-inactivated blood (100 mL) were combined. Using a pipette, eight ml of the mix was put into a 50ml culture flask and 32 ml of the mix into a 200ml culture flask, and the slant was allowed to settle. The media was then kept at 40°C until it was needed (Tankeshwar 2016).

Sodium chloride 4.5g, potassium chloride 0.2g, calcium chloride 0.1g, sodium bicarbonate 0.1g, and D-glucose (anhydrous) 1.25g are the constituents in Locke's solution (all from Sigma). All the materials were weighed, combined, and dissolved in 500 mL of distilled water to make the solution. The preparation was then autoclaved for 30 minutes at 121°C. To inhibit the growth of bacteria and fungi, L-Glutamine (0.29 mg/ml) and penicillin-streptomycin (100IU/ml-100g/ml) were added to the autoclaved Locke's solution. The pereparation was then filtered with a 0.2m pore size diameter syringe filter and kept at 40°C until needed (Tankeshwar 2016).

3.3.7.3. Parasite culture

The skin scrapings of the specimen were injected in duplicate into the NNN medium with Locke's solution as an overlay at the research locations to culture promastigotes. After that, the cultures were incubated at 240°C. Cultures were examined under an inverted microscope (Leitz-Wetzlar, Germany) starting on the fourth day after inoculation and at three-day intervals subsequently to observe the development of Leishmania promastigotes. If samples were found to be contaminated by bacteria or other organisms of no interest, they were discarded right away. Negative cultures were sub-cultured into new NNN media after two weeks, and those cultures that remained negative after two weeks were judged negative and discarded (Aberra et al. 2019). All positive cultures were subcultured in a liquid medium containing RPMI 1640 medium (Sigma), 10% heat-inactivated bovine calf serum (Sigma), 2mM glutamine, 100 units/ml penicillin, and 100g/ml streptomycin (GIBCO BRI, Scotland), and grown to stationary phase as required for subsequent tests (Tasew, Kebede, Wolday, et al. 2009:2).

3.3.7.4. DNA extraction from cultured samples

During the late exponential growth phase of Leishmania promastigotes, they were harvested. The phenol-chloroform and ethanol precipitation procedure were used to isolate the genomic DNA from the promastigotes (Sambrook & Russell, 2000). Promastigotes were pelleted by centrifuging at 3,000 rpm for 15 minutes (Beckman Coulter, Allgera 6R centrifuge, USA), then washed twice in Phospate Buffer Saline at 3,000 rpm for 10 minutes and transferred to a 1.5 ml Eppendorf tube. 300 liters of lysis buffer were used to break down the parasites (10mM Tris-Cl, pH 8.3, 50 mM EDTA, 1% SDS). The lysate was then incubated for 1 hour at 370C with 20g RNase (Pharmacia) at a final concentration of ten l/ml to destroy contaminating RNA, and then overnight at 420C with 20g Proteinase K (GIBCO) at a final concentration of ten l/ml to remove proteins that would interfere with further DNA analysis. Then, with a proportion of 24,24, and one, respectively, phenol, chlorofom, and isoamyl were added to each sample and vortexed for five seconds (Aberra et al. 2019).

After centrifugation at 12,000xg for five minutes at room temperature, the top phase was gently separated and transferred to 1.5 ml Eppendorf tubes. 350 mL isopropanol was added to each sample, which was then kept at -20^{0}C for 30 minutes. The samples were then centrifuged at 1,2000xg for 15 minutes. After removing the isopropanol, 1 mL of ice-cold 70% ethanol was added. After five minutes of incubation at -20^{0}C, the samples were centrifuged for five minutes. The ethanol was withdrawn from the DNA pellets, and they were left to dry at room temperature. To dissolve the dried pellet, it was re-suspended in 20l DNase/RNase free 1X TE buffer (pH = 8.0) and heated for 10 minutes in a 68^{0}C water bath. The concentration of DNA was determined spectrometrically at the absorbance of 260nm using 50-fold dilutions, and 1g/l dilutions were prepared. The DNA samples were kept at -20°C until they were further processed (Aberra et al. 2019).

3.3.7.5. Polymerase chain reaction

Temperatures for denaturation, annealing, and elongation were optimised in all PCR experiments. Each PCR reaction contained 50 l of LITSR/ L5.8S primers and

25 l of 13A/13B and Laer3Lash/ Laef3Lash primers. In all PCR experiments, a hot start master mix (QIAGEN GmbH, Germany) was used. In all cases, a mix devoid of template DNA was used as a PCR internal control. 35, 40, and 32 PCR cycles were used for LITSR/ L5.8S, 13A/13B, and Laer3Lash/ Laef3Lash, respectively. In a 1.8% agarose (Sigma) gel with 0.5 g/ml ethidium bromide, the PCR products were seen. Each PCR product was placed onto the gel in four microliters with one liter of 5x loading buffers (0.25% Bromophenol blue, 0.25% Xylene cyanol FF, and 50% Glycerol). Three microliters of 0.1g/l molecular size markers were injected as a marker. At 100V and 50mA, a 90-minute electrophoresis split was performed. The photographs were taken with a UV trans-illuminator (UVP-imager) (in Epi-chemi II, Upland, with a camera linked to it and a computer) (Bugssa et al. 2014; Bsrat et al. 2015).

3.3.7.6. Identification of the species using ITS1-PCR- RFLP method

For species identification, the internal transcribed spacer (ITS-1) in the ribosomal operon was amplified with primers LITR/L5.8S and restriction digestion with enzymes Hha I. The restriction enzymes Hha I were used to digest the ITS-1 amplicon. The reaction mix contains 17 liters of PCR product, 2 liters of 10x enzyme buffer, and 1 liter (10 U) of enzyme separately. The reaction mixture was vortexed before being incubated at 37°C for two hours. For band size measurement, the restriction fragments were analysed in 2% agarose (Sigma) in the presence of 0.5g/mlethidium bromide and molecular size markers. On the gel, ten liters of the sample were mixed with 2.5 liters of 5x loading buffers. To estimate the size of DNA fragments, a molecular size marker of four microliters (0.1g/l) was loaded. Electrophoretic separation was carried out for 90 minutes at 100 V and 50 mA, with pictures collected and used for further analysis (Bugssa et al. 2014; Bsrat et al. 2015; Mohammadiha, Dalimi, Mahmoodi, et al. 2017:385-386; Monroy-Ostria, Nasereddin, Monteon, Guzmán-Bracho & Jaffe 2014:2).

3.3.8. Data analysis

3.3.8.1. Knowledge attitude and practice survey data analysis

Dependent variables in this section are knowledge attitude and practice and independent variables are demographic characters (sex, age, education, religion, marital level, and type of residential area). Percentage, frequency, and mean values were used to descriptively summarise the data.

The association between demographic characteristics and an overall score of knowledge, attitude, and practice of the participants about CL was investigated using Pearson's chi-square test of association. For categorical variables, the chi-square is employed to determine statistical significance. With categorical data for two independent variables, Pearson's chi-square test of association is used to examine if there is any relationship between the variables (Suresh 2019).

To investigate independent variables that could potentially predict the probability of a higher level of overall knowledge, attitude, and practice of study participants about CL, a binary logistic regression analysis (both bivariate with crude odds ratio and multivariate with adjusted odds ratio) at 95% confidence intervals (CI) was performed. Accordingly, predictors of the probability of high levels of various subscales of knowledge, attitude, and practice of CL were also identified using logistic regression. Regression evaluation is a type of predictive modeling method that is applied to find the connection between a dependent and independent variable. Multiple regression is used when the independent variables used to predict or explain the outcome of the dependent variable are two or more (Pedamkar 2020).

The application of logistic regression is proper when the dependant variable is dichotomous or has a binary outcome. The three types of logistic regression are. binary logistic regression where the categorical response has only two likely results (Example: Yes or No), Multinomial logistic regression is with three or more unordered categories (Example: predicting who will be USA president in 2020) and

ordinal logistic regression in which three or more ordered categories are involved (Example: Movie rating from 1 to 5) (Swaminathan 2018).

Since the dependent variable in this survey is dichotomous (satisfactory /unsatisfactory knowledge, favorable /unfavorable attitude, and good/poor practice), binary logistic regression was found to be the proper statistical tool. After calculating crude odds ratio and adjusted odds ratio, significant level was set if p-value is less than 0.05. Tables as well as bar graphs were used to present results and data were computerised and analysed using SPSS for windows version 24.

3.3.8.2. Risk assessment survey analysis

CL positive (either with an active lesion or scars) was the dependent variable in the risk assessment survey, whereas the independent variables included socio-demographics such as age, sex, knowledge, dwelling location, and associated household, host, animal, and environmental factors.

To summarise the given variables in demographic characters, residential futures, illness characters, and household, environmental, host, and animal elements, descriptive analysis with frequency, mean, and percentage, as well as descriptive bar graphs, pi-charts, and tables were employed.

Bivariate logistic regression was used to calculate crude odds ratio and every independent variable was treated with the dependent variable separately. For calculation of adjusted odds ratio in multivariate logistic regression, a backward stepwise method was used. Stepwise logistic regression helps to decrease the number of predictor variables for building the best logistic regression model by choosing key variables to get a simple and clearly explainable model. There are two methods of stepwise logistic regression forward and backward stepwise selection (Choueiry 2020).

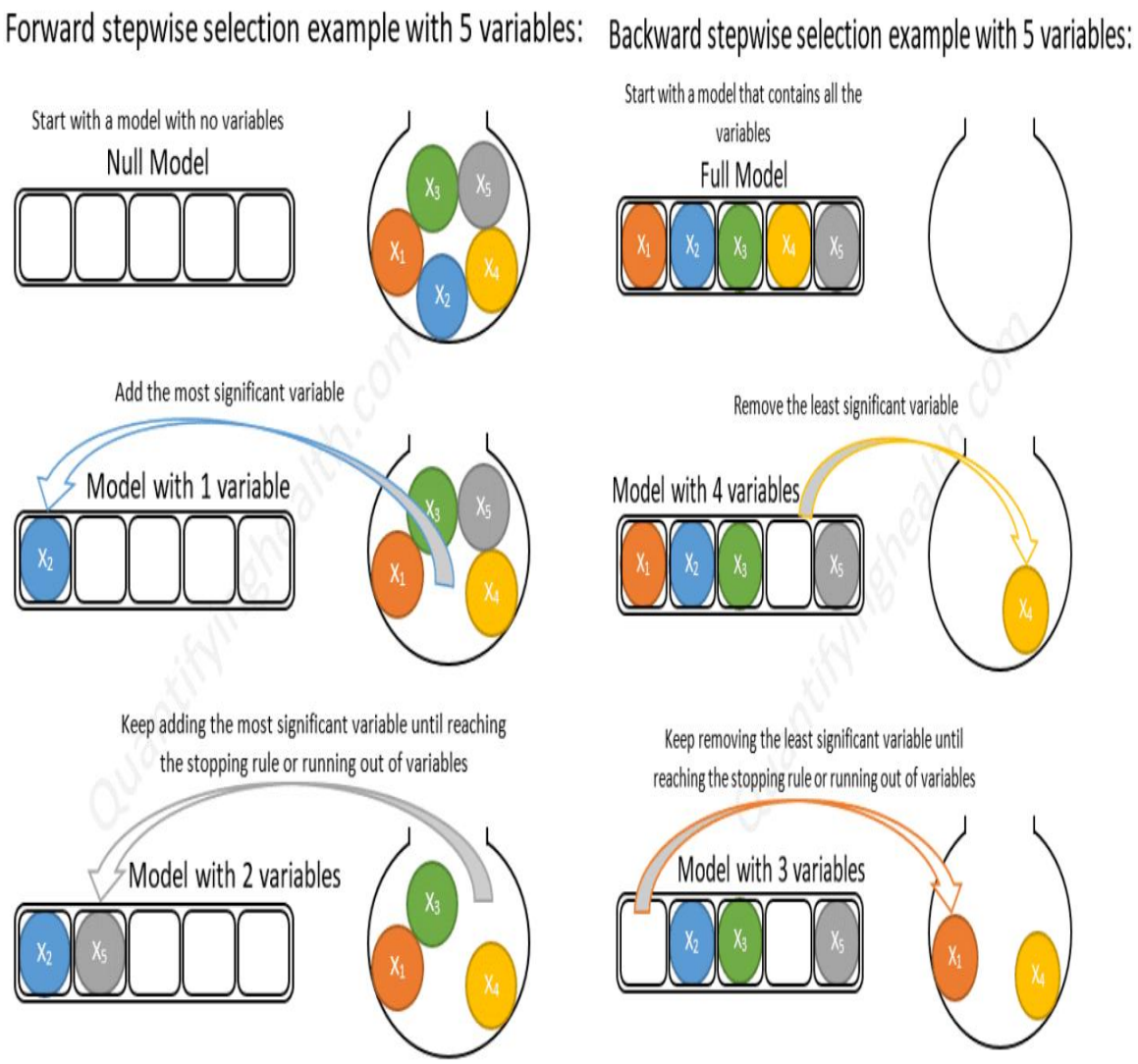


Figure 3.3: Diagram of steps in forward and backward stepwise (Choueiry 2020).

In forward stepwise selection, the analysis starts with the null model without variable and continues with inserting the very significant variables one following the other. On the contrary, with backward stepwise selection, the starting model is a full model containing all variables under consideration, then will go on deleting the slightly significant variables one after the other. In both cases addition or removal of variables to the model is stopped at a pre-specified stopping rule is achieved or when all variables are tested. The advantage of backward stepwise selection over the other is that it allows consideration of the impacts of all variables at the same

time. This is particularly significant when the variables in a model are interrelated, as backward stepwise may be required to maintain them all in the model. Furthermore, unless the number of candidate variables exceeds the sample size (or the number of occurrences), a backward stepwise procedure is recommended (Choueiry 2020). As a result, the majority of predictive factors in this study were chosen using a backward stepwise strategy, with statistical significance set at $p < 0.05$. R Package Version 3.02 was used to conduct the statistical analysis.

Multiple Correspondence Analysis (MCA), as an Exploratory Data Analysis (EDA) procedure that recognises patterns in big and complicated datasets was also applied to identify key factors contributing to the transmission of CL. MCA is a technique that permits examining the relationship between two or more qualitative variables and produces maps where it is possible to visually explore distanc between the categories of the qualitative variables and between the observations (Husson & Josse 2008:163). MCA graphical overviews make representation of the interactions between variables easier without the requirement of any prerequisites, thus making understanding simpler (Jalayer & Zhou 2016). The MCA analysis was done using R software (Version 3.03) and FactoMineR package.

Finally, hierarchical cluster analysis was done for identifying similar groups of data in a dataset. If groups share the same characters they will be clustered together, therefore, with clustering, the population will be divided into groups that are similar. Thus, groups with similar traits will be clustered (Kassambara and Fabian 2020). There are various kinds of clustering techniques such as model-based clustering, comprising partitioning systems, fuzzy clustering, hierarchical clustering and density-based clustering (Pedamkar 2020). Hierarchical clustering is a tactic for pinpointing groups in the dataset which provides a tree-based description of the objects called a dendrogram. Observations are converted into groups sharing similar traits by cutting the dendrogram at the desired similarity level (Bock 2020).

3.3.8.3. Prevalence survey analysis

Here frequency and percentages tables were used for analysis and presentation of results. Bi-variate logistic regression with 95% level of confidence was applied for evaluate identifying correlation of herbal treatment application and positivity in smear and culture test. This test was done using a package in R Package Version 3.02 and p-value of < 0.05 was considered statistically significant.

3.3.9. Ethical considerations

The Research and Ethics Committee, Department of Health Studies at UNISA, granted ethical permission for the study with the number REC-012714-039/HSHDC/457/2015 (Annexure F). Unisa Ethiopia Learning Centre wrote a letter to SNNPR Health Bureau expressing support for the study's goals (Annexure G). The protocol for this study was approved by the Department of Zoonotic Disease Unit at Addis Ababa University Akililu Lemma Institute of Pathobiology (ALIPB), which was part of the thematic "studies on major vector-borne parasitic diseases of people including zoonoses in chosen areas in Ethiopia." Support letters had been obtained from the SNNPR Health Bureau to conduct study at Sodo Woreda Health Bureau (Annexure I).

Each study's participants were fully informed about the study's purpose and objectives. The right to engage or not participate in study activities was also communicated to participants. By avoiding the recording of personal identifiers, the participants' confidentiality and data security were protected throughout the study duration (such as name). Data collectors working with participant information were required to sign a data use and confidentially agreement. Owing to the exclusion of personal identifiers during recording and entry/upload to databases, any data shared with researchers within the project was by default be de-identified.

The research purpose was fully stated to the applicants prior to obtaining their consent, and all adults who participated in the study gave their informed consent (Annexure J). An assent form was prepared and submitted to children who are

under 18 years old. The paranets of these children were also asked if they are willing of their children are involved in the study (Annexure J). The consent and assent forms include information on the aim, procedures, and potential harms and benefits from participating in the research. Individuals were also asked if they are willing if their pictures (Part of the lesion) were taken and used for publication. All suspected CL cases received a free laboratory diagnosis and referral to treatment. The study used only samples from willing individuals, and the results were kept private.

3.4. INTERNAL AND EXTERNAL VALIDITY OF THE STUDY

Internal and external validity can vouch for the study's credibility and significance. Internal validity is concerned with the study's framework as well as its execution. External validity focuses on the applicability of the research findings (Cuncic 2020). This study was performed with the utmost consideration to keep the standards of validity and reliability. Thorough attempts were made to make sure that the design, sampling methods, tools, and techniques of the research in adaptation of sound principles attain internal and external validity of data as well as their reliability in generating equivalent findings if employed in similar settings.

3.4.1. Validity and reliability

Validity the measure of capacity of survey tool or an equipment to provide precise results that meet expectations (Polit & Beck 2010:377). The validity of the research tools such as questionnaires were examined to meet standards by preparation of relevant research questions and then by pretesting those before commencement of the study. The questionnaire was created by searching many literatures as well as the researcher's own experience in order for the tool to capture the entire content domain. In addition, to assess the validity of tool content, an expert on the KAP survey from ALIBP Addis Ababa University and other colleagues who are experts in cross-sectional surveys reviewed the tools (Polit & Beck 2010:378).

Reliability is a concept that deals with ability of an instrument to provide similar results or outcomes in repeated trials. Reliable research is the one with methods and procedures that supply identical results if applied at another time (Polit & Beck, 2010:373). In this research, standardised questionnaire was prepared to achive reliability. The questioners were pilot tested on similar set of populations before the actual study was started. After this the tools were properly modified according to the input obtained during pre-testing. Pretested questions were compared if they provide similar findings with the main study.

3.4.2. Internal validity

Internal validity ascertains that conclusion about cause-and-effect relationship drawn from the research are reliable and trustworthy (Polit & Beck, 2010:373). Internal validity of this study was maintained by designing research questions that are in line with study objectives. In addition, participants were randomly selected on voluntary bease. Dependant and independent variables were identified by reviewing litratures. The researcher's supervisors inspected each item for its correctness to the research questions. The respondents were drawn from the six kebeles and included individuals from randomly sampled households. The data gathering tool was pretested with 30 individuals who did not join the main study.

3.4.3. External validity

After setting a sampling frame of households in each of the six kebeles which are distributed proportionaly based on the entire population, households were randomly selected by use of simple random sampling thecniques. In this study, a comprehensive explanation of the study technique, statistical analysis and summarisation of the results was conducted in a transparent manner. The raw data obtained from the three KAP survey, risk assessment and prevalence study are kept for interested parties and will be attached as a supplementary document in journals in during future publication. For conformability, academic and ethical faithfulness were kept by the researcher. Notes, personal identifyers, and other sensitive issues were kept secured.

In general, actions comprising development of proper tools, effective training of the data collectors, use of a standardised questionnaire, tight supervision of all field activities, cautious data entry, and use of appropriate data analysis methods were executed to confirm data truthfulness. Sampling procedures, data collection guidelines and setting of excluding and including standards of this study are up to the level of keeping strong internal and external validity.

3.5. CONCLUSION OF METHODOLOGY

The research strategy, the expressive and exploratory nature of the study, the data collecting, and analytic methodologies, validity, and ethical considerations of the study were all covered in this chapter. The data from the KAP survey and the risk assessment survey were analysed using descriptive statistics, univariate analysis, and logistic regression. Multiple correspondence analysis and clustering approaches were also used to discover factors that played a significant role in the spread of CL in the research area. The research was carried out in six purposively chosen kebeles in the Sodo District in Southern Ethiopia. Administration of structured questionnaire interviews, clinical sample collection, and laboratory diagnoses were used for data collection. The findings of this study will be explained in the next chapter.

CHAPTER 4

ANALYSIS, PRESENTATION AND DESCRIPTION OF THE RESEARCH FINDINGS

The techniques for collecting data and the designs employed to achieve the research objectives were discussed in the previous chapter. The KAP survey, the risk assessment survey, and the species identification study are all described in this chapter, together with their analysis, presentation, and description.

4.1. PHASE ONE- ANALYSIS OF KNOWLEDGE ATTITUDE AND PRACTICE SURVEY FINDINGS

This study has different phases. The first phase is the study of knowledge, attitude, and prevention practice of the Sodo community on CL. The second one deals with an investigation of the disease prevalence in the district and detection of the parasite species existing in human hosts.

4.1.1. Data management and analysis

The data analysis plan was given due attention from the beginning. Inspections were performed all through the data collection processes for data validity, consistency, and completeness. The KAP study strategy used in this research certifies that comparable methods can be certainly done in a related placing. The analysis includes the delivery of structured interviews to 423 household heads randomly selected from six kebeles by trained health extension workers living and employed by health office in the district. These extension workers are well familiar with the residential area, social norms, and health status as they work being assigned in a specific kebele with certain number of households.

Upon the end of the data gathering activity, thorough data entry templates were designed in SPSS version 24.0 and all data from the structured questionnaire were effectively read into the software by the principal investigator. Statistical analysis of the data was performed from July to October 2017. Data clean-up, explanatory and

inferential statistical examination were done. This study employed some appropriate statistical assessment and measurement standards including descriptive statistics, bivariate and multivariate logistic regression methodologies.

4.1.2. Research results of knowledge attitude and practice survey

The questionnaire containing of structured questions was created in such a way that it achieves evidence on socio-demographic futures, knowledge on CL, attitude scale towards CL, and prevention practice related to CL. Knowledge scores were classified as unsatisfactory if the score is < 4 and while if it is > 4 then it was set to be satisfactory knowledge. Attitude scores between 0 and 3 were put as unfavorable but scores > 3 categorised as having favorable. Practice score of 4 and above were stared as good prevention practices.

4.1.2.1. Socio-demographic characteristics

A total of 423 individuals were involved in this study; 219 (51.8%) of the participants were males and 204 (48.2%) were females. One hundred fifty (35.5%) were in the age range of 34.5 to 44.5 years and 107 (25.3%) are in the age group of 24.5–34.5. Orthodox 391 (92.4%) was the dominant religion in Sodo District. Participant's major occupation was farming [216 (51.1%)] and 188 (44.4%) of the participants claim to be housewives. The average family size within the study participants was 5.76 and the average duration of stay in the district was 32.79 years. Accordingly, only a few proportions of the study participants 81 (19.1%) came from another place while the rest 342 (80.9%) are originally from the same district.

Considering the education level of the respondents, 216 (51.1%) were illiterates while 119 (28.1%) of the participants can read and write only. None of them travelled out of their district in six months of the study time. Of the 351 (82.8%) participants who claim to have farming land, the majority 218 (51.5%) confirmed the land is situated near their home. Few 27 (6.4%) have farming land near a river and in the case of 106 (25.1%) of the study participants, the farm is found both near

home and near a river. The general socio-demographic features of the respondents were presented in Table 4.1.

Table 4.1: Demographic characteristics of the respondents (N=423).

Characteristics	Frequency	Percent
Gender		
Male	219	51.8
Female	204	48.2
Age		
18.0-24.5	24	5.7
24.5–34.5	107	25.3
34.5- 44.5	150	35.5
44.5-54.5	82	19.4
Over 54.5	60	14.2
Religion		
Orthodox	391	92.4
Protestant	29	6.9
Muslim	3	0.7
Educational level		
Illiterate (cannot read & write)	216	51.1
Can read and write (Only)	119	28.1
Primary education (1-6 grade)	63	14.9
Secondary education (8-12 grade) & above	25	5.9
Occupation		
Farmer	216	51.1
Housewives	188	44.4
Trader	19	4.5
If yes location of farmland?		
Near home	218	51.5
Near a river	27	6.4
Both	106	25.1
other	72	17.0
Place of birth		
Same district	342	80.9
Another place	81	19.1

4.1.2.2. Description of knowledge of cutaneous leishmaniasis and factors affecting knowledge

In this section, knowledge of the community is presented concerning responses provided for knowledge measuring questions. In addition, socio-demographic factors affecting knowledge are described employing the chi-square test and logistic regression.

4.1.2.2.1. Knowledge of the community about cutaneous leishmaniasis

Knowledge of CL among study participants is presented in Table 4.2. Two hundred and sixteen (51.1%) of the participants were able to identify the disease as CL after they were shown a picture illustrating cases of CL reported from different parts of Ethiopia (Annexure K) and 350 (82.7%) of the participants have heard about CL. This may indicate that many of the participants who were unable to identify the disease may hear about it but lack the proper knowledge about the disease. However, the majority of 350 (82.7%) of the participants have heard about CL mostly being informed by families, friends, neighbours, and colleagues 286 (67.5%) and from being exposed to the disease 38 (9.0%). Professionals like health workers, religious leaders, teachers, and teaching tools like media, brochures, and newsletters which are believed to be vital gear for community health awareness creation programmes collectively share an exceedingly small percentage 38 (9.0%) of the knowledge attained about the disease.

Table 4. 2: Knowledge of CL among study participants (N=423)

Variables (Multiple responses are possible)		**Frequency**	**Percentage**
Heard about CL	Yes	350	82.7
	No	73	17.2
Major Cause of CL	Do not know	226	53.4
	Germ	52	12.3
	Other	46	10.9
	Drinking unclean water	44	10.4
Sign and symptoms	Fever	56	13.2
	Plaque	268	63.4
	Skin rash	56	13.2
	Pus containing	206	48.7
	Papule	55	13.2
	Do not know	92	21.7
How is the disease transmitted?	Mosquito bite	30	7.1
	Contact with patient	24	5.7
	Sandfly bite	52	12.3
	Do not know	252	59.4
Biting and blood-sucking behaviour of the vector	Aware	210	49.6
	Not aware	198	46.8
Knowledge (overall)	Satisfactory	263	61.9
	Unsatisfactory	161	38.1

When asked if they know anybody who is infected by the disease in their area, most 301 (71.2%) of the respondents knew someone infected with CL. Of these, 218 (51.5%) knew that the person they know to have CL was infected in the past and 80 (18.9%) claim that the person they know is actively infected. According to the respondents in the current study, the individuals they know of being infected are neighbours, family members, cousin, and self in 171 (40.4%), 52 (12.3%), 25 (5.9%), and 21 (5.0%) of the respondents respectively (Figure 4.1). Multiple responses of 10 (2.4%), 8 (1.9%), 8 (1.9%) and 3 (0.7%) were also recorded for self and neighbour, family member and neighbour, family member and self and finally for family member/self/ and neighbour, respectively.

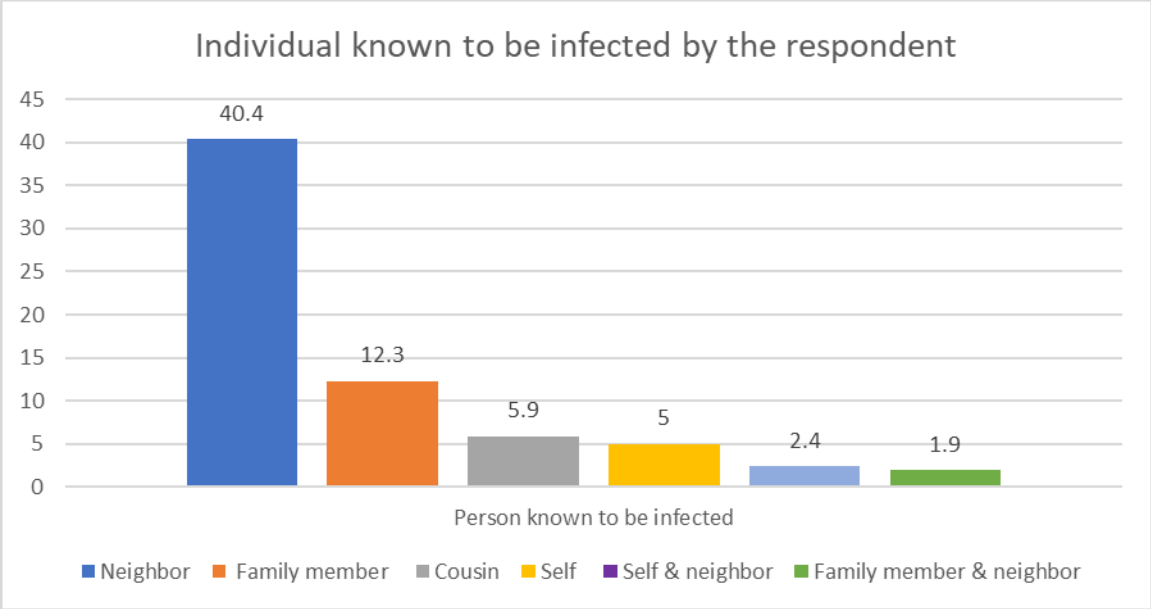


Figure 4.1: Relationship of persons known to be infected with CL and respondent.

Knowledge of the cause of CL and its transmission means is vital for the prevention of the disease. Rural communities with limited access to awareness creation programmes may have a wrong perception of the cause of the disease as being religions, a sign of sinfulness, or a curse. If people have a wrong perception about the cause of the disease, then they will tend to be reluctant in preventing themselves from sandfly bit since they are not fully aware of the cause and

transmission means of the disease. Two hundred six (53.4%) of the respondents claim that they do not know the cause of CL, 52 (12.3%) mentioned germ, and 44 (10.4%) say drinking unclean water could cause the disease. More than half 252 (59.4%) of the study participants are not aware on how the disease is transmitted while only a small proportion of the study population (52 (12.35%)) knew the disease is communicated by a bite of sandfly vector.

Any objective proof of CL, such as a skin rash is a sign and this can help family members, and the individual experiencing the signs to identify the disease and seek medical attention. The current study revealed that plaque and pus-containing wounds were mentioned as the main clinical symptoms of CL by 268 (63.4%) and 206 (48.6%) of the respondents respectively while 92 (21.7%) admitted that they did not know any symptom of CL.

Nose (316 (74.5%)), face (305 (71.9%)), forehead (116 (27.4%)) and ear (102 (24.1%)) are parts of the body mentioned by the respondents to be mostly infected by the lesion (called "kuselet" in Amharic) from CL infection.

Anti-leishmaniasis vaccines are not available for human. Travellers to endemic areas will be most at risk and are recommended wear proper clothes with long sleeves if possible impregnated with repllents and avoid outdoor activities at night. In addition, these travlers must use insect repellent such as DEET on uncovered skin, sleeping under insecticide-impregnated bed nets or in a room with fan to disturb sandfly movement through ventilation, and sleeping in raised position from the floor as sandflies are weak fliers (Fit for travel, 2020). In this study, only 93 (21.9%) knew that there is a possibility of acquiring leishmaniasis by travelling to areas that are prone to the disease.

In addition to transmitting leishmaniasis, sandflies can also serve to transmit other diseases such as Carrión's disease, sandfly fever, vesicular stomatitis, summer meningitis and Chandipura virus encephalitis (Dvorak, Halada, Hlavackova, et al., 2014:1). Proper knowledge about the vector plays a role in actions against

preventing leishmaniasis and other diseases. To measure the knowledge of study participants about the sandfly vector, a picture of the vector that is believed to exist in the southern part of Ethiopia was shown to the respondents and their reply was recorded (Annexure K). As a result, nearly half 210 (49.6%) reported to know the biting and blood-sucking behaviour of phletomine sandfly.

Sandfly eggs are placed in moist, dark locations like livestock barns and poultry. Sanitation by removing bushes and vegetation, fixing cracks and crevices in the wall and floor, and separating cow barns and poultry from human residences are thus additional vector control measures in addition to insecticides (Adam et al. 2017:15&16). Awareness where sandflies breed and rest are important so that the community in endemic areas could undertake serious measures of sanitation. In the current study, 123 (29.0%) and 139 (32.8%) of the participants do not know the breeding place and the preferred biting time of the vector, respectively. But 178 (42%) said that the vector bits mostly at dusk. Dirty places 218 (51.5%), water ponds 194 (45.9%), and garbage collection sites 111 (26.2%) are the most mentioned breeding sites of the vector. A little percentage of response was given to cervices in the house 18 (4.3%), thatched roof 17 (4.0%), and cattle shades 20 (4.7%).

4.1.2.2.2. Factors affecting knowledge of the study participants

Looking into the overall scores of knowledges in the current study, 262 (61.9%) of the respondents have a satisfactory general knowledge of CL. A chi-square statistic was used to investigate the factors that affect the knowledge of the respondent (Table 4.3).

Considering gender as one of the predictor variables overall knowledge scores was satisfactory in 152 (74.5%) of female respondents and 110 (50.2%) of the male respondents. This result is statistically significant (X^2=26.414, P 0.000). In our study, a satisfactory level of overall knowledge was observed in illiterate individuals

(n=141, 65.2%) and able to read and write (n=57, 47.8%) which is statistically significant (X^2=24.373, P<0.001).

Table 4.3: Distribution of satisfactory CL knowledge of participants (N=423).

Characteristics	Total sample N=423 No (%)	No (%) Satisfactory Knowledge	X^2	*p-value*
Gender			26.414	0.000*
Male	219(51.8)	110(50.2)		
Female	204(48.2)	152(74.5)		
Age			8.341	0.080
18.0-24.5	24(5.7)	15(62.5)		
24.5–34.5	107(25.3)	74(69.1)		
34.5- 44.5	150(35.5)	94(62.6)		
44.5-54.5	82(19.4)	51(62.1)		
Over 54.5	60(14.2)	28(46.6)		
Educational level			24.373	0.000*
Illiterate	216(51.1)	141(65.2)		
Can write & read (Only)	119(28.1)	57(47.8)		
Primary (1-6 grade)	63(14.9)	52(82.5)		
Secondary & above	25(5.9)	12(48.0)		
Occupation			25.683	0.000*
Farmer	216(51.1)	109(50.0)		
Trader	19(4.5)	12(63.1)		
Housewife	188(44.4)	141(75.0)		
Place of birth			5.050	0.025*
Same district	342(80.9)	203(59.3)		
Other place	81(19.1)	59(72.8)		
Know a person with CL infection			67.962	0.000*
Yes	301(71.2)	223(74.0)		
No	120(28.4)	37(30.8)		
Overall knowledge				
Satisfactory		261(61.9)		
Unsatisfactory		161(38.1)		

*p < 0.05 (significant)

Two hundred three (59.3%) of the respondents who are originally from Sodo District and 59 (72.8%) who are not born in the district, have satisfactory CL knowledge which is statistically significant (X^2=5.050, P 0.025). Similarly, statistical significance (X^2=25.683, P= 0.000) was observed in the respondents with a different occupation where higher percentages of satisfactory knowledge were recorded in 141 (75.0%) of housewives. In the current study, respondents who know a person infected with

CL have a higher percentage of satisfactory knowledge (74.0%) of the disease. This is statistically significant (X^2=67.962, P 0.000).

For evaluation and to analyse the effect of selected variables on participants' knowledge, attitude, and practice of CL, binary logistic regression (for both bivariate and multivariate analysis) was utilised (Table 4.4). The result of the bi-variate analysis reveals a statistically significant relationship between gender (p< 0.000), education level (p< 0.002 & p< 0.011), occupation (p< 0.000), place of birth (P< 0.026), and knowledge of a person with CL (p< 0.000) and CL knowledge of the participant. Hence, females are knowledgeable than males [COR=2.9; 95% CI, 1.919-4.371]. Accordingly, females are three times likely to have satisfactory knowledge than males.

Table 4.4: Comparison of demographic variables with knowledge of CL (N=423).

Variables		***COR (95% CI)***	***AOR (95%CI)***
Gender			
	Male	1.00	1.00
	Female	2.897(1.919-4.371)	1.997(0.640-6.233)
Age			
	18.0-24.5	1.00	1.00
	24.5–34.5	1.345(0.535-3.385)	0.951(0.339-2.666)
	34.5- 44.5	1.007(0.414-2.453)	0.875(0.317-2.416)
	44.5-54.5	0.987(0.386-2.525)	1.077(0.367-3.157)
	Over 54.5	0.525(0.199-1.384)	0.656(0.212-2.030)
Educational level	Illiterate	1.00	1.00
	Able to write & read	0.489(0.310-0.771)	0.838(0.470-1.494)
	Primary	2.515(1.238-5.105)	3.879(1.729-8.703)
	Secondary & above	0.491(0.213-1.130)	0.613(0.238-1.580)
Occupation			
	Farmer	1.00	1.00
	Trader	1.683(0.638-4.437)	1.039(0.296-3.645)
	Housewife	2.945(1.926-4.502)	1.527(0.477-4.891)
Place of birth			
	Same district	1.00	1.00
	Another place	1.836(1.075-3.136)	0.750(0.369-1.524)
Know a person with CL infection	Yes	1.00	1.00
	No	0.156(0.098-0.248)	0.163(0.099-0.268)

Respondents who can read and write are less likely to be knowledgeable compared to those who are illiterate [COR=0.49; 95% CI, 0.310-0.771]. This means there is no significant difference between illiterates and who can read and write for acquiring knowledge of CL. On the contrary, respondents who receive primary education are more likely to have a better knowledge of CL than those who are illiterate [COR=2.5; 95% CI, 1.238-5.105].

Individuals who are not originally from the Sodo District are knowledgeable compared to participants who come from other places [COR=1.84; 95% CI, 1.075-3.136]. Study participants who claim not to know a person infected with the disease are less knowledgeable than those who do [COR=0.156; 95% CI, 0.098-0.248].

Multivariate analysis also shows a statistically significant association between educational level, previous knowledge of a person with CL and level of knowledge. Respondents receiving primary education [AOR=3.879; 95% CI, 1.729-8.703] are found to be knowledgeable than respondents who are illiterates. This shows that for education to be a factor affecting knowledge, the level must be at least to receiving a primary level education. Those who do not know someone infected with CL are less knowledgeable than those who do [AOR=0.163; 95% CI, 0.099-0.268]. From the results in the chi-square test and logistic regression, it can generally be concluded that after adjusting for all variables, receiving primary education, and knowing someone infected with CL are factors that truly contribute to satisfactory knowledge of CL.

4.1.2.3. Attitude towards cutaneous leishmaniasis and factors affecting it

Answers provided for the six questions which are used to measure attitude are explained here and these are compared to other research findings. Finally, socio-demographic factors were identified using the chi-square test and logistic regression.

4.1.2.3.1. Attitude towards cutaneous leishmaniasis in Sodo District

The attitude of the study participants in the area is recorded in Table 4.5. Two hundred and sixty-one (61.7%) of the participants in this study believed that CL can be treated. This attitude is important for seeking treatment and reducing the disease prevalence. However, 70 (16.5%) of the members percived that CL is not curable and 85 (20.1%) have no idea whether the disease can be treated or not.

Table 4.5: Attitude of the Sodo community towards CL (N=423).

Variables (Multiple responses are possible)		**Frequency**	**Percentage**
Do you think CL can be treated?	Yes	261	61.7
	No	70	16.5
	I have no idea	85	20.1
What is your preferred choice of medication for CL?	Modern medicine	142	33.6
	Herbal medicine	251	59.3
	I do not know	28	6.6
What do you think is the outcome of CL if untreated	Death	28	6.6
	Disfiguring	264	62.4
	Self-cure	1	0.2
	Both (death & disfigure)	67	15.8
	I have no idea	54	12.8
	Other	8	1.9
Do you think that you are well informed about CL?	Yes	167	39.5
	No	248	58.6
	Other	7	1.6
Willing to Participated in CL control programs?	Yes	328	77.5
	No	82	19.4
	Other	12	2.8
How serious is CL in your community?	Very serious	273	64.5
	Ordinary	31	7.3
	Not very serious	46	10.9
	I do not know	66	15.6
	Other	6	1.4
Attitude (overall)	favorable	226	53.4
	unfavorable	197	46.6

The importance of hygiene in the living/sleeping area was indicated to be very important for the prevention of the disease as indicated by a recently published research paper (Younis, Kroeger, Joshi, er al. 2020:11). Partly 257 (60.6%) of the

participants in this study share the fact that environmental hygiene is important for the prevention of CL transmission.

More than half 251 (59.3%) of the respondents preferred indigenous medication through the application of herbs by traditional healers.

In a qualitative study done in the USA, three main types of explanations for escaping medical care were discovered (Taber, Leyva & Persoskie 2015:29). These were unfavorable opinions of requesting medical treatment, low perceived need to seek medical care over expectation that the illness will improve in timen, cost of treatment, absence insurance, and time limits (Taber, Leyva & Persoskie 2015:29). In the current study, of those who do not seek modern medication claim lack of trust in modern medication 165 (38.9%), trusting herbal medication 143 (33.7%) high cost of modern medication 95 (22.4%) and not having time to attend medical care because of being busy with work 69 (16.3%) as the main reasons for not seeking modern treatment (Figure 4.2).

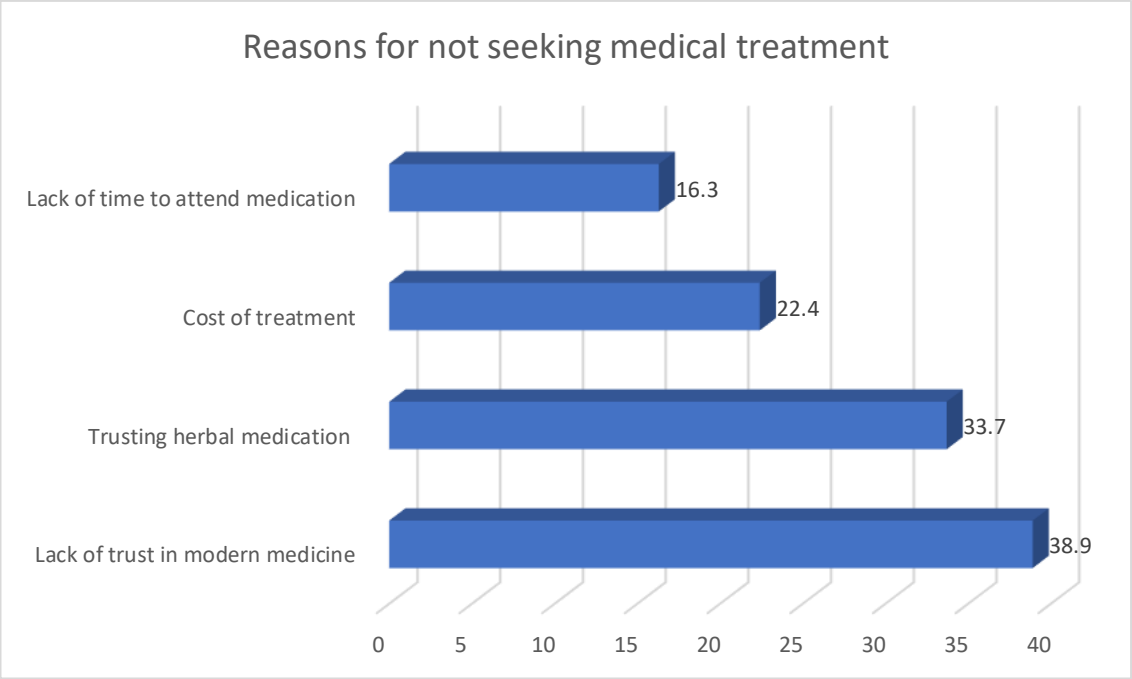


Figure 4.2: Reasons for not seeking medical treatment.

In this study, lack of awareness 171 (40.3%), insufficient budget 114 (26.9%), and dependency on herbal medicine 99 (23.3%) were major constraints mentioned by the informants for not being able to control the disease within the community. Lack of such awareness creation programmes can also be explained by the fact 248 (58.6%) of the respondent confess that they do not think they are well informed about CL. Never having a chance to learn about the disease and inadequate source of information in the locality were mentioned by 203 (47.9%) of the respondents as reasons for not being well informed about the disease.

In this study, majority (64.6%, n=274) see CL as a very serious problem in their locality. Generally, 226 (53.2%) of the participants have a favourable attitude while 197 (46.4%) have unfavourable attitudes.

4.1.2.3.2. Factors affecting the attitude of the community towards cutaneous leishmaniasis

Dissemination of favourable attitude towards CL among the study sample using the chi-square test is listed in Table 4.6. Females show a higher frequency 124 (59.3%) record of favorable attitude which is statistically significant (X^2=5.486, p 0.019). In this study, a favourable attitude towards CL was observed in 90(60.0%), 60(56.0%), and 43 (52.4%) of the respondents who are in age groups of 34.5 to 44.5, 24.5 to 34.5, and 44.5 to 54.5 respectively. This result was statistically significant (X^2=10.168, p 0.038).

One hundred and seventy-three (50.5%) and 53 (65.4%) of the respondents originally from Sodo District and those from other have favourable attitude which was statistically significant (X^2 = 5.802, p 0.016). One hundred ninety-three (64.1%) respondents who know CL infected person have favorable attitude which is statistically significant (X^2 = 46.272, p 0.000).

In bivariate analysis, age (p< 0.048 and P< 0.018), occupation (p<0.028), place of birth (p< 0.017), and knowing CL infected person (p< 0.000) were seen significantly

associated with attitude (Table 4.7). Therefore, those with age group of 24.5-34.5[COR=2.553; 95% CI, 1.007-6.475] and 34.5- 44.5 [COR= 3.000; 95% CI, 1.208-7.448] have a favourable attitude about CL compared to those in the 18.0-24.5 age group.

Table 4.6: Distribution of favorable attitude towards CL (N=423).

Variables	**Total sample N=423 No (%)**	**No (%) Favorable attitude**	***X²***	***p-value***
Gender			5.486	0.019*
Male	219(51.8)	105(47.9)		
Female	204(48.2)	121(59.3)		
Age			10.168	0.038*
18.0-24.5	24(5.7)	8(33.3)		
24.5–34.5	107(25.3)	60(56.0)		
34.5- 44.5	150(35.5)	90(60.0)		
44.5-54.5	82(19.4)	43(52.4)		
Over 54.5	60(14.2)	25(41.6)		
Educational level			3.279	0.351
Illiterate	216(51.1)	108(50.0)		
Able to write & read (Only)	119(28.1)	64(53.7)		
Primary (1-6 grade)	63(14.9)	39(61.9)		
Secondary & above	25(5.9)	15(60.0)		
Occupation			5.149	0.076
Farmer	216(51.1)	105(48.6)		
Trader	19(4.5)	9(47.3)		
Housewife	188(44.4)	112(59.5)		
Place of birth			5.802	0.016*
Same district	342(80.9)	173(50.5)		
Another place	81(19.1)	53(65.4)		
Know a person with CL infection			46.272	0.000*
Yes	301(71.2%)	193(64.1)		
No	120(28.4%)	33(27.5)		

*$p < 0.05$ (significant)

The multiple logistic regression analysis shows that respondents who are in the 34.5 to 44.5 age group [AOR= 3.584; 95% CI, 1.299-9.890] and who can read and write [AOR= 1.929; 95% CI, 1.095-3.399] have favourable attitude than those who are in 18.0-24.5 age group and who are illiterate.

Table 4.7: Comparison of socio-demographic variables with the attitude to CL (N=423).

Variables		*COR (95% CI)*	*AOR (95%CI)*
Gender			
	Male	1.00	1.00
	Female	1.583(1.077-2.326)	1.278(0.455-3.588)
Age			
	18.0-24.5	1.00	1.00
	24.5–34.5	2.553(1.007-6.475)	2.569(0.930-7.093)
	34.5- 44.5	3.000(1.208-7.448)	3.584(1.299-9.890)
	44.5-54.5	2.205(0.850-5.719)	2.587(0.905-7.400)
	Over 54.5	1.429(0.530-3.852)	2.359(0.726-7.305)
Educational level			
	Illiterate	1.00	1.00
	Able to write &read	1.164(0.743-1.822)	1.929(1.095-3.399)
	Primary	1.625(0.915-2.886)	1.895(0.980-3.663)
	Secondary & above	1.500(0.645-3.487)	2.371(0.924-6.083)
Occupation			
	Farmer	1.00	1.00
	Trader	0.951(0.372-2.434)	0.844(0.259-2.744)
	Housewife	1.558(1.050-2.313)	1.345(0.470-3.847)
Place of birth			
	Same district	1.00	1.00
	Another place	0.541(0.327- 0.896)	1.613(0.851-3.056)
Know a person with CL infection	Yes	1.00	1.00
	No	0.212(0.133-0.338)	0.221(0.136-0.358)

4.1.2.4. *Prevention practice and factors regarding cutaneous leishmaniasis*

Actions performed by the community to manage, control, and prevent the disease are described here. Findings are discussed and compared to reports from similar studies. Factors identifying individuals as having poor or good practices were also identified.

4.1.2.4.1. Prevention practice of Sodo community regarding cutaneous leishmaniasis

The practice of the Sodo community against CL was summarised (Table 4.8). In most cases, CL heals by itself over time but if it is MCL, it can lead to disablity and tissue damage. Where species identification is impossible, treatment is based on

expert opinion (Vries et al. 2015:104). When asked how CL was treated, the application of herbal remedies 312 (73.8%) and the use of specific drugs at health centre 187 (44.2%) were mentioned more frequently. Only one person (0.2%) suggested home rest without medication.

CL, being vector born disease with no vaccination and clearly defined treatment guideline, is difficult to control. Therefore, prevention is better than therapy for both the patient as well as affected communities. About half 235 (55.6%) of participants in this study use preventive measures. The preventive measures used by participants in this study are bed nets 178 (42.1%), cleanliness 118 (27.9%), early detection and treatment 114 (27.0%), and insecticide 94 (22.2%) for prevention of the disease (Figure 4.3). Poor prevention practice was observed on personal protective measures (wearing covering clothes 5 (1.2%), use of insect repellents 28 (6.6%) which are important as a first-line defence mechanism against sandfly bite.

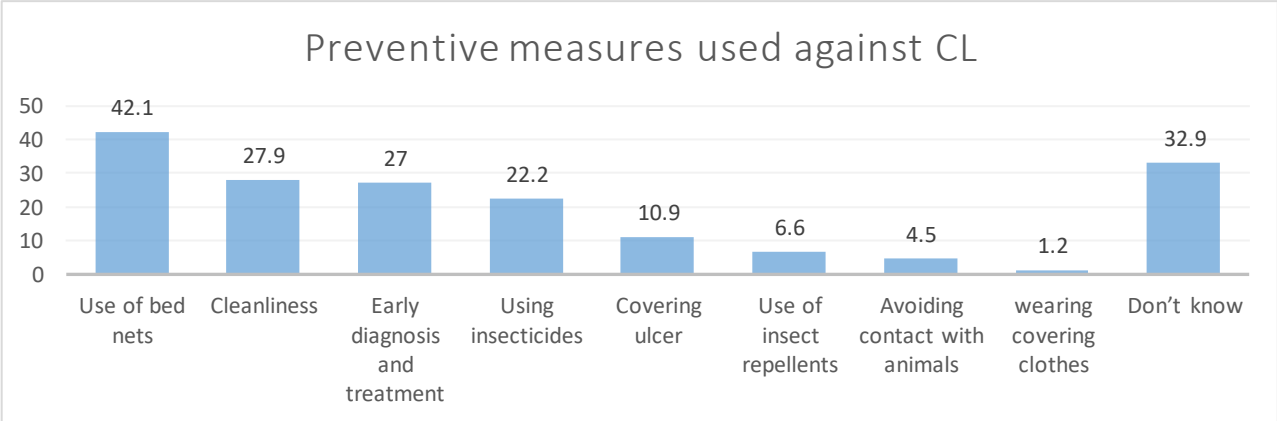


Figure 4.3: Type of major CL preventive measures used in Sodo District.

The presence of bed nets in the household does not guarantee the prevention of the disease unless properly used while sleeping. For maximum impact, nets must be used consistently and correctly. In a study conducted at southern Ethiopia, it was stated that knowledge of household insecticide treated nets (ITNs) utlilization is vital in addition to estimating how many households have bed nets. This was suggested because there was underutilisation of available ITNs at a household plus vulnerable

members of the household were not primary users (Berkessa, Oljira & Tesfa 2016:2).

Table 4.8: Practice of the Sodo community against CL (N=423).

Variables (multiple responses)		**Frequency**	**Percentage**
How CL was treated	Herbal	312	73.8
	Home rest without	1	0.2
	medication	31	7.3
	Praying	187	44.2
	drugs given at health center	13	3.1
	Vaccination	27	6.4
	Holy water	53	12.5
	I do not know		
Use preventable measure	Yes	235	55.6
	No	130	30.7
	I do not know	44	10.4
Having bed net in the house	Yes	288	68.1
	No	124	29.3
	Do not know	11	2.6
Sleeping outside or spending time outside	Yes	19	4.5
	No	392	92.7
Using bed net when sleeping	Yes	251	59.3
	No	135	31.9
	Do not know	36	8.5
Using repellents	Yes	128	30.3
	No	244	57.7
	Do not know	51	12.0
Work time preference at high T^o	Day time	148	35.0
	Night	37	8.7
	Both	165	39.0
	Very early morning	44	10.4
Practice (overall)	Good	213	50.4
	Poor	201	46.9

In this study, it was found that 251 (59.3%) use bed nets when they are sleeping. Different studies in Ethiopia reported that there is non-utilisation of available nets among different communities range from 21.5% to 73.3% (Teklemariam, Awoke, Dessie & Weldegebreal 2015:3). Accordning to the national plan to prevent and control malaria, all households in malaria endemic area should have at least two insecticide treated bed nets to decrsease mortality due to malaria to 75% by 2020 (Ministry of Health – Ethiopia 2020). Looking into the number of bed nets in the

current study, of those who use bed nets 105 (24.8%) and 69 (16.3%) have a maximum of two or three bed nets in the house, respectively. The coverage of bed net in the area is not enough and will probably not be effective for the prevention of leishmaniasis as sandflies are much smaller than mosquitos, so a tightly woven net is needed.

Spending time outside at night or sleeping outdoors could expose persons to sandfly bite. This type of practice is quite common in the northern part of Ethiopia owing to large farms where male workers move to and sleep outdoor to escape peak temperatures. In this study, only 19 (4.5%) have a custom of spending time outside or sleeping outdoors.

More than half (57.7%) of the respondents do not use a repellent. Any type of insecticide has never been sprayed in 210 (49.5%) of the participants' houses. Of the 170 (40.0%) insecticide sprayed houses, spraying was for protection against malaria. In Ethiopia, malaria and leishmaniasis, though may co-exist in the same places, are distributed in different geographical areas. Hence, control programmes intended for malaria may or may not control leishmaniasis.

When asked about their preferred time of work at peak temperature, the respondents prefer to work at daytime 148 (35.0%) and both at daytime and night (39.0%). Generally, 210 (49.4%) of the respondent have good prevention practice while 214 (50.4%) of the respondents have poor practice.

4.1.2.4.2. Factors affecting the practice of cutaneous leishmaniasis

Table 4.9 shows results of chi-square test. Good practice was observed in 125 (61.2%) of the female, 15 (62.5%), and 65 (60.7%) of the 18.0-24.5 and 24.5–34.5 age group, 40 (63.4%) of the primary education, 118 (62.7%) of the housewives. This observation was statistically significant at (X^2=18.795, p 0.000) for sex, (X^2=13.569, p 0.009) for age, (X^2=10.039, p 0.012) for level of education and (X^2=21.251, p 0.000) for occupation.

Good practice was seen in 160 (37.8%) of the respondents who originally came from another district and 53 (65.4%) of those who happen to know someone infected with CL which is statistically significant at (X^2=9.110, P 0.003) and (X^2=28.477, P 0.000) respectively.

Table 4.9: Association between demographic characters and practice scores (N=423).

Characteristics	**Total sample N=423 No (%)**	**No (%) Good practice score**	***X²***	***p-value***
Gender			18.795	0.000*
Male	219(51.8)	88(40.1)		
Female	204(48.2)	125(61.2)		
Age			13.569	0.009*
18.0-24.5	24(5.7)	15(62.5)		
24.5–34.5	107(25.3)	65(60.7)		
34.5- 44.5	150(35.5)	68(45.3)		
44.5-54.5	82(19.4)	44(53.6)		
Over 54.5	60(14.2)	21(35.0)		
Educational level			10.039	0.012*
Illiterate	216(51.1)	114(52.7)		
Able to write & read	119(28.1)	48(40.3)		
Primary	63(14.9)	40(63.4)		
Secondary & above	25(5.9)	11(44.0)		
Occupation			21.251	0.000*
Farmer	216(51.1)	86(39.8)		
Trader	19(4.5)	9(47.3)		
Housewife	188(44.4)	118(62.7)		
Place of birth			9.110	0.003*
Same district	342(80.9)	160(46.7)		
Another place	81(19.1)	53(65.4)		
Know someone with CL			28.477	0.000*
Yes	301(71.2%)	117(38.8)		
No	120(28.4%)	36(30.0)		

*p < 0.05 (significant)

Table 4.10 presents the result of logistic regression of demographic variables and overall practice score. Accordingly, in bivariate logistic regression female sex (p< 0.000), over 54 age group (p<0.024), being able to read and write (p< 0.030), housewives (p< 0.000), coming from other districts (p<0.003) and knowing

someone with the diseases (p<0.000) are significantly associated with good practice.

Table 4.10: Socio-demographic characters and practice of participants (N=423).

Variables		*COR (95% CI)*	*AOR (95%CI)*
Gender			
	Male	1.00	1.00
	Female	2.355(1.594- 3.480)	0.841(0.309-2.292)
Age			
	18.0-24.5	1.00	1.00
	24.5–34.5	0.929(0.373-2.314)	0.768(0.293- 2.019)
	34.5- 44.5	0.498(0.205- 1.208)	0.453(0.174- 1.181)
	44.5-54.5	0.695(0.273- 1.767)	0.703(0.258- 1.914)
	Over 54.5	0.323(0.121- 0.863)	0.359(0.123- 1.048)
Educational level	Illiterate	1.00	1.00
	Able to write & read	0.605(0.384-0.952)	0.852(0.493- 1.475)
	Primary	1.556(0.873- 2.774)	1.797(0.938- 3.444)
	Secondary & above	0.703(0.305- 1.628)	0.776(0.309-1.948)
Occupation			
	Farmer	1.00	1.00
	Trader	1.360(0.531- 3.486)	1.345(0.423-4.277)
	Housewife	2.548(1.705- 3.809)	2.142(0.768- 5.972)
Place of birth			
	Same district	1.00	1.00
	Another place	2.153(1.300- 3.567)	1.335(0.718- 2.483)
Knowing someone with CL	Yes	1.00	1.00
	No	0.300(0.191- 0.472)	0.315(0.195- 0.510)

As a result, female [COR=1.5; 95% CI, 1.031- 2.233], those who can read and write [COR=1.5; 95% CI, 1.031- 2.233], housewives [COR=1.5; 95% CI, 1.031- 2.233], those who originally come from other districts [COR=1.5; 95% CI, 1.031- 2.233] and those who know someone infected with the disease have good practice than men. In contrast, illiterates, farmers, those who are born in Sodo and those who previously did not meet with a person infected with the disease.

In multiple logistic regression only knowing someone infected with the disease show statistical significance relation with good practice regarding CL [OR=0.315; 95% CI, 0.195- 0.510)].

4.1.3. Overview of KAP research finding

4.1.3.1. Overview of knowledge of study participants about cutaneous leishmaniasis

Knowledge was assessed using an 8-item questionnaire including

- If they could name the disease after being shown a picture of CL manifestation.
- Have you ever heard of CL?
- What is the cause of CL?
- What are the major symptoms of CL?
- How is the disease transmitted?
- Whether they know the importance of the vector as a transmitter of leishmaniasis after being shown a picture of phletomine sandfly believed to exist in area.
- Where do sandflies breed?
- When is the preferred biting time of the vector?

Regarding knowledge status and gaps of CL, the following were observed

- Nearly half, 207 (48.9%) of the respondents are not able to identify a case of leishmaniasis when shown a picture of the case, though the majority, 350 (82.7%) have heard about it.
- The cause is not known to more than half 226 (53.4%) of the respondents.
- Symptoms of the disease and body parts affected by the lesion (such as nose, face, forehead, and ear) were well known among the communities.
- Transmission means of the disease was not properly understood by the community as a bite of sandfly was known by 12.3% (52/423) of the respondents and 59.4% (252/423) of them do not know how it is transmitted.
- The sandfly vector was identified as an "important biting and blood-sucking insect" upon seeing a picture of the fly by 210 (49.6%) of the population.

- The community has unsatisfactory knowledge about the preferred biting time of the vector as only 178 (41.0%) said it bite at dusk and 139 (32.8%) do not know the biting time.
- One hundred and twenty-three (29.0%) of the respondents do not know the breeding place of the vector plus little percentage of response was given to cervices in the house 18 (4.3%), thatched roof 17 (4.0%) and cattle shades 20 (4.7%).

Analysis of the relationship between the demographic factors and knowledge of the disease shows that:

- Gender, level of education, place of birth, occupation, and knowledge of a person infected with CL were statistically significant factors determining knowledge status in the chi-square test.
- Gender, education level, occupation, place of birth, and knowledge of a person with CL and CL knowledge of the participant were shown to have a statistically significant association in bivariate logistic regression.
- There is a statistically significant correlation between educational level, previous knowledge of a person with CL, and level of knowledge in Multivariate logistic regression.

4.1.3.2. Overview of Sodo community attitude towards cutaneous leishmaniasis

The attitude was evaluated based on a 6-point questionnaire:

- Do you think chewie can be treated?
- What is your preferred choice of medication for the treatment of Chewie?
- What is your opinion on outcome of CL left untreated?
- Do you feel that you are well informed about chewie?
- Willingness to participate in CL control activities
- Is CL a serious health problem in your locality?

Overview of the community attitude is summarised as:

- The treatable nature of the disease is well recognised among the community as 261 (61.7%) confirm that it can be treated. However, the preferred choice of medication was the application of traditionally known herbs for 251 (59.3%) of the population.
- More than half 264 (62.4%) of the communities correctly think that the disease outcome is a disfiguring effect.
- Lack of disease information to the community is justified by the fact that 248 (58.6%) of the respondents did not feel well informed about it.
- There is a willingness among the majority 328 (77.5%) of the study population to take part in CL control programmes if there is such activity in the area.
- CL is considered a serious disease by 273 (64.5%) of the participants.

An analysis of the relationship between the demographic factors and attitude towards CL explains:

- Gender, age, place of birth, and knowledge of someone with CL were associated with attitude in the chi-square test.
- Age, occupation, place of birth, and knowing someone with CL have a significant association with the attitude of the community in bivariate analysis.
- The presence of an association between attitude and age, level of education, and knowledge of someone with CL was seen in multivariate logistic regression.

4.1.3.3. Overview of practice of the community

Prevention practice had eight items:

- How was chewie treated?
- Use of preventable action for CL?
- Type of methods used to prevent CL.
- Having bed nets in the house,
- Sleeping condition and
- using bed nets when sleeping.
- Using repellents and
- Work time preference when the temperature is peak.

General outputs from answers to these questions are the following:

- Treatment practice taken by majority 312 (73.8%) of the community was the application of herbal remedies. Low health service utilisation for CL was evident as only 187 (44.2%) sought treatment from health facilities.
- More than half 235 (55.6%) of the participants practice the use of preventive measures against the disease.
- Use of bed nets (42.1%), cleanliness (27.9%), early detection and treatment (27.0%), and application of insecticides (22.2%) were widely used methods for preventing the disease among the communities. However, the use of personal protective measures such as wearing covering clothes and the use of insect repellents are less practiced.
- A sufficient proportion of the community 288 (68.1%) has bed nets in the house. However, the number of participants using bed net while sleeping is lower than 251 (59.3%).
- Sleeping condition practiced by the community is encouraging as only 19 (4.5%) sleep outside or have a habit of spending time outside at night.

- The use of repellents was mentioned by only 128 (30.3%) of the respondents.
- Working time preference in some community (39.0%) day and night, 10.4% early mornings, and 8.7% at night) pose exposure to the bite of sandfly vector.

The relationship between the demographic factors and preventive practice of the community indicates:

- Gender, age, level of education, know someone with CL, and practice are related in the chi-square test.
- Significant association of practice with gender, age, level of education, occupation, place of origin, and knowing someone with CL in bivariate logistic regression.
- Only knowing someone infected with the disease having statistical significance relation practice regarding CL.

4.1.3.4. Overview of overall knowledge attitude and practice

Regarding the overall knowledge of the Sodo community about CL, 263 (61.9%) have satisfactory knowledge of CL while 161 (38.1%) have unsatisfactory knowledge. The percentages of study participants showing favorable and unfavorable attitudes about CL are 53.4% (n=226) and 46.6% (n=197) respectively. About half 213 (50.4%) of the participants exhibit good preventive practice against the disease while the rest 210 (49.6%) have a poor practice of CL (Figure 4.4).

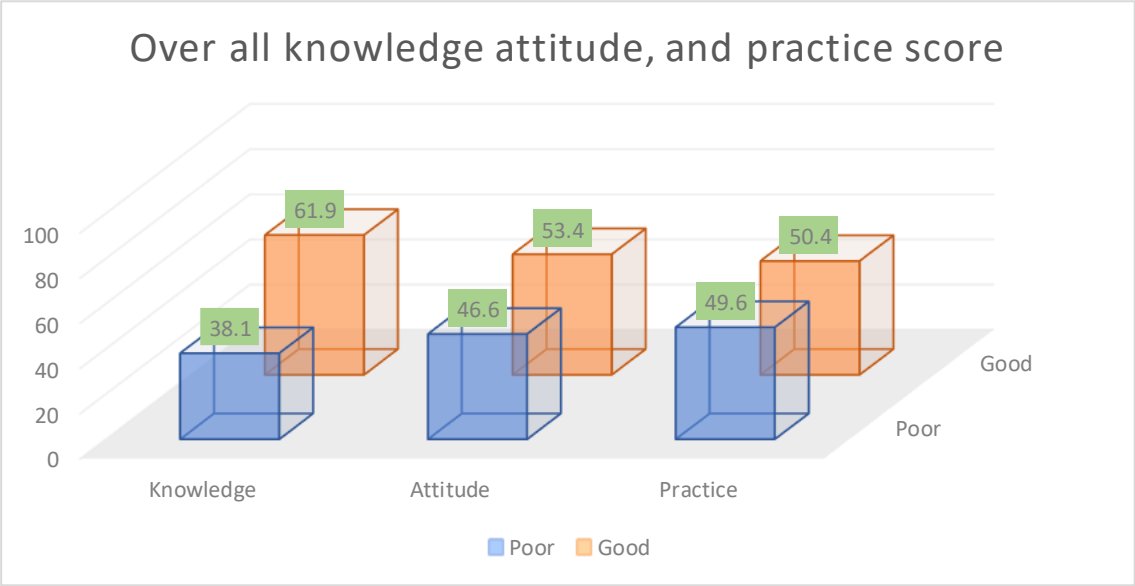


Figure 4.4: Overall KAP score of the community.

Comparing overall practice with knowledge and attitude in the current study (Table 4.11), the findings indicate that 262 (61.9%) respondents with satisfactory knowledge 173 (40.9%) have a good practice. The association between knowledge and practice was statistically significant at p<0.000. Accordingly, the odds of having a good practice are nearly five times higher for those who have satisfactory knowledge than those who have unsatisfactory knowledge.

Table 4.11: Comparison of overall knowledge, attitude, and practice (N=423).

	PRACTICE		**Total** No (%)	**p- value**	***COR (95% CI)***
	Poor No (%)	Good No (%)			
Knowledge					
Unsatisfactory	121(28.6)	40(9.5)	161(38.1)	1	
Satisfactory	89(21.0)	173(40.9)	262(61.9)	0.000	4.722(2.992 - 7.454)
Total	210(49.6)	213(50.4)	423(100)		
Attitude					
Unfavourable	134(31.7)	63(14.9)	197(46.6)	1	
Favourable	76(18.0)	150(35.5)	226(53.4)	0.000	3.213(2.081 - 4.960)
Total	210(49.6)	213(50.4)	423(100)		

A similar relationship was seen between attitude and practice as of the 226 (53.4%) participants who have favorable attitude, 150 (35.5%) also have a good practice. This was also statistically significant at p<0.000 with individuals with favourable attitude having three times more likely to have a good practice.

4.2. PHASE TWO-ASSESSMENT OF FACTORS AFFECTING CUTANEOUS LEISHMANIASIS

The second phase of the research focuses on the identification of risk factors. Households were selected using a systematic method and the household head was asked about the family members with active infection or about past infection. Household factors (wall type and condition, floor and roof type, presence of latrine, and screen) were recorded by observation of the health extension worker delivering the questionnaire. Socio-demographic, residential features, environmental conditions, host factors, and animal factors were assessed by interviewing everyone within the house. Then if a member of the household was found to be actively infected with CL skin slit samples were taken for microscopic examination with Giemsa staining and culture. The results of the clinical sample analysis will be presented in phase three (Prevalence study). The identification and analysis of different risk factors playing a role in the transmission dynamics of the disease within the community are described next.

4.2.1. Data management and analysis

Variables were classified into different groups for analysis as socio-demographic factors, residential features, household factors, environmental factors, host factors, and animal factors. Descriptive analysis was done by measuring and comparing frequency and percentage. In addition, logistic regression both bivariate and multivariate were used to assess the association of these factors to the occurrence of the disease Data were cleaned and some variables were manipulated to avoid assumptions of logistic regression.

Variable manipulation was done by:

A. Screening predictors based on descriptive statistics.

Before beginning to develop a model, it is critical to gain a full understanding of the data. Descriptive statistics (means, variance, percentiles, and so on for continuous variables, and frequency tabulations for categorical variables) can be useful in identifying variables that aren't important in the model. As a result, it is preferable to maintain variables that have been accurately and precisely measured, as well as

variables that are somewhat complete. The following specific guidelines were applied to select variables:

- Avoiding variables with a large number of missing observations.
- Select only variables with substantial variability (e.g., if almost all participants are male, adding gender as a predictor is not likely helpful).
- If a categorical variable has many categories with small numbers of observations in each, consider combining categories (if this makes a biological sense), or eliminate the variable (Dohoo, Martin & Stryhn 2014:369).

B. Correlation analysis

When all pairwise correlations between predictor variables are looked at, pairs of variables with essentially the same information are found. Multicollinearity occurs when highly correlated variables are included in a model, which can result in unstable coefficient estimates and erroneous standard errors. If two highly correlated variables are found, one should be chosen for inclusion in the model based on biological plausibility, fewer missing observations, measurement convenience, and/or measurement reliability (Dohoo, Martin & Stryhn 2014:369).

Details of procedures done to manipulate variables are explained in this chapter under the factor analysis section.

4.2.2. Research findings of the risk assessment study

In this section, different factors contributing to the transmission of CL and their association with the occurrence of the diseases will be discussed.

4.2.2.1. Descriptive analysis of the study population and disease factors

The population living in six kebeles of Sodo woreda was studied as described in Chapter 3 section 3.3.3.2. A total of 379 households and 1,356 individuals (all individuals within a given household) agreed to participate in the study. All were included, and clinical assessment was done for individuals with a suspected case of leishmaniasis.

4.2.2.1.1. Socio-demographic features

Social and demographic features of the participants are presented in Table 4. 12. Average family size was 5.5 (with min=1 and max=11). Regarding the marital status of the respondents, the majority (60.47%) were married and the rest 39.52% were not married. Six hundred ninety-five (51.2%) of the respondents were female and the rest 661 (48.5%) were male. The majority of the respondents are in the age group of 10-17 years old 306 (22.5%), 35-44 years 295 (21.76%), and 25-34 years 259 (19.1%).

Almost all 1,290 (95.13%) of the study participants are followers of Orthodox religion. More than 98.0.% (1330) of respondents did not obtain any education or are educated up to secondary school. Considering the occupation of the study population 619 (45.6%) have farm-related work and the rest 739 (54.4%) are engaged in non-farm related work of these 462 (62.54%) and 253 (34.29%) were students and had no formal work, respectively. One hundred twenty-three (9.07%) of the participants were infected with the parasite. Of these, 49 (3.61%) were with active infection during the visit, and 74 (5.45%) were infected in the past.

Table 4.12: Demographic characteristics of the community (N=1356).

Characteristics	Frequency (%)
Age	
5-10	10(0.74)
10-17	306(22.57)
18-24	183(13.50)
25-34	259(19.10)
35-44	295(21.76)
45-54	172(12.68)
>54	121(8.92)
Gender	
Female	695(51.25)
Male	661(48.75)
Religion	
Muslim	16(1.18)
Orthodox	1290(95.13)
Protestant	50(3.69)
Education	
Unable to read and write	454(33.48)
Able to read and write	185(13.64)
Primary education	422(31.12)
Secondary education	269(19.84)
Higher education	5(0.37)
Informal education	21(1.55)
Occupation related to farm	
Yes	619(45.65)
No	739(54.35)
Occupation (other than farming) (N=739)	
Students	462(62.54)
No formal job	253(34.29)
Government employee	12(1.59)
Children	8(1.10)
Service provider	4(0.54)
Infection status	
Positive	123(9.07)
Negative	1233(90.93)
Time of infection	
Active	49(3.61)
Past	74(5.46)

4.2.2.1.2. Description of residential features

Adazer kebele comprises the largest households (37.54%) visited during the study. As explained in chapter 3, this was proportionally done by considering the total number of households. Beki is the smallest kebele of the six only with a sample of 77 households. Almost all (92.26%) of participants are born and raised in Sodo District while few (7.74%) move to and make a living in the district. Of those respondents who have a farm or whose livelihood depends on the farm, the farm location is near home (29.3%), both near home/river (17.7%) and near a river (3.17%). This shows that half 50% (681/1356) of the respondents have or work on a farm which is located near home or a river (Table 4.13).

Table 4.13: Residential and working features of study populations (N=1356).

Characteristics	Frequency (%)
Residence Kebeles	
Adazer	509(37.54)
Beki	71(5.24)
Genete Mariyam	326(24.04)
Kela Zuria	171(12.61)
Kola Nurena	98(7.23)
Michael Semero	181(13.35)
Born and raised in same district?	
Yes	1,251(92.26)
No	105(7.74)
Farm location	
Near home and near river	240(17.70)
Near river	43(3.17)
Near home	398(29.35)
Not near home or river	88(6.49)
No farm	587(43.29)

4.2.2.1.3. Household factors

Sodo District is a rural area, but the centre (Kela Zuria kebele) can be referred to as having an urban setting relative to the other kebeles in the district. House building styles in these rural areas follow the same design. The walls are usually made of wood and mud. Some may add to this plastering with cow dung. The roof will be covered with locally grown and dried grasses or with steel depending on the household's economic status. In addition, the floor will be made from mud and

sometimes like on the wall; the floor is plastered with cow dung as a beautification and cleaning strategy. These typical houses are called "*Gojo bet*" and they are homes for many people in rural areas of Ethiopia (Figure 4.5).

Figure 4.5: Typical house in the study area called *"Gojo bet"* (JoeCaz 2006).

The interviewer recorded variables measuring household factors after observing conditions in and around the house. Therefore, the walls were found to be either plastered with cow dung (56.5%) or plastered with mud (42.7%). Both types of houses are made of mud but there is the additional step of plastering the wall with cow dung in houses plastered with cow dung while in the other kind the wall is left with mud. Almost all houses (376) except for the three made from stone and bricks are made of mud. In rural areas of Ethiopia, cow dung is used for painting the wall and floor of the house and this is done regularly at a few months interval or when there is a family occasion or public holidays. The presence of organic matters in the household attracts sandflies and increases the risk of exposure to their bite (Fateh, Vatandoost, Rassi, et al. 2019:7).

Looking into the wall condition, cracks or holes were not observed in the majority of the houses 289 (76.3%). Wall conditions in nearly one fourth 90 (23.7%) of the households represents the presence of risky wall condition (holes formed (1.3%), cracked (10.6%) and with both crack and holes (11.8%) creating favourable vector hiding places in the house which in turn will increase the exposure of household members to vector bite and consequently disease transmission. Of these houses

with cracked or holes, it was some of the walls with cracks/holes in the majority (49.38%) of the houses and another 14.28% of houses have either cracks or holes on almost all walls. The roof was made of steel in the majority (70.1%, n=266) of the houses in the study area.

The 355 (93.7%) houses have a functional latrine. The interviewing health extension worker was asked to grade the condition of the latrine according to the standard set by the district health office. Accordingly, 21.4% were leveled as "good", 57.8% as "Fair" and the rest 14.5% as having "bad" condition. Open-ground toilets can be an ideal breeding place for sandflies because they are usually surrounded by a wet floor. Moreover, individuals who go to the restroom at dusk during the main vector biting time may be subjected to infective bites (Younis et al. 2020:10).

Table 4.14: Description of house condition (N=379).

Variables	Frequency (%)
Wall type	
Plastered with cow dung	214(56.46)
Plastered with mud	162(42.74)
Other (stone/bricks)	3(0.79)
Wall condition	
Cracked & holes formed	45(11.87)
Holes formed	5(1.32)
No cracks/holes	289(76.25)
cracked	40(10.55)
Roof (Celling) type	
Steel	266(70.18)
Dry grass (Straw)	108(28.50)
Other (stone/made of fabrics)	5(1.32)
Floor type	
Cemented	7(1.85)
Plastered with cow dung	345(91.03)
Mud	27(6.07)
Presence of functional latrine	
Absent	24(6.33)
Present	355(93.66)
Presence of screen on window/door	
Yes	88(23.22)
No	291(76.68)

Of houses in which health extension workers were able to observe the surrounding area, 185 (13.67%) and 112 (8.28%) of the houses are built around near creek/waterways and pits, respectively. Areas of this type will favour the creation of microenvironments for breeding and resting of the sandfly vectors.

Putting screens on the window or door denies entry of sandflies into the house minimises the risk of exposure to the household members (Jacquelyn**,** 2018). Screens on window or door were found only in 88 (23.2%) of the houses. Based on the interviewer's (health extension worker) observation, 37.0% of the screens were in good condition and 13.67% were not in a good condition while the rest were unevaluated.

4.2.2.1.4. Environmental factors

Different environmental factors are known affect leishmaniasis. Acacia and sensel trees (*Adathada shimperina)* may serve as a resting place for the sandfly and may increase the exposure of individuals to sandfly bite (Pareyn, Kochora, Rooy, et al. 2020:2). Half (52.95%) and 21.98% of the household have acacia and sensel tree around the house, respectively.

Different environmental factors ease human- vector contacts such as the presence of backyard with trees, farm, and cave, or gorges in which reservoir hosts may live. Nearly a quarter (23.23%) of the respondents live in a place near to the presence of a backyard and a field. Only 13.94% of residents say that they have a public assembly area near their home. The majority (72.94%) of the respondents in the households confirm the existence of a farm within 300m radios. The presence of cave or gorge in the area was 48.60%, 38.79%, and 23.75% in villages, in the working area, and around the household area, respectively.

Water sources for individuals living in the study area are tap water (39.0%), river (28.0%), hand-dug well (23.0%), and spring (10.0%) (Figure 4.6). Adding the three water sources except tap water makes 61.0% of the contribution. This means that most of the individuals live near water sources or are in close contact with these.

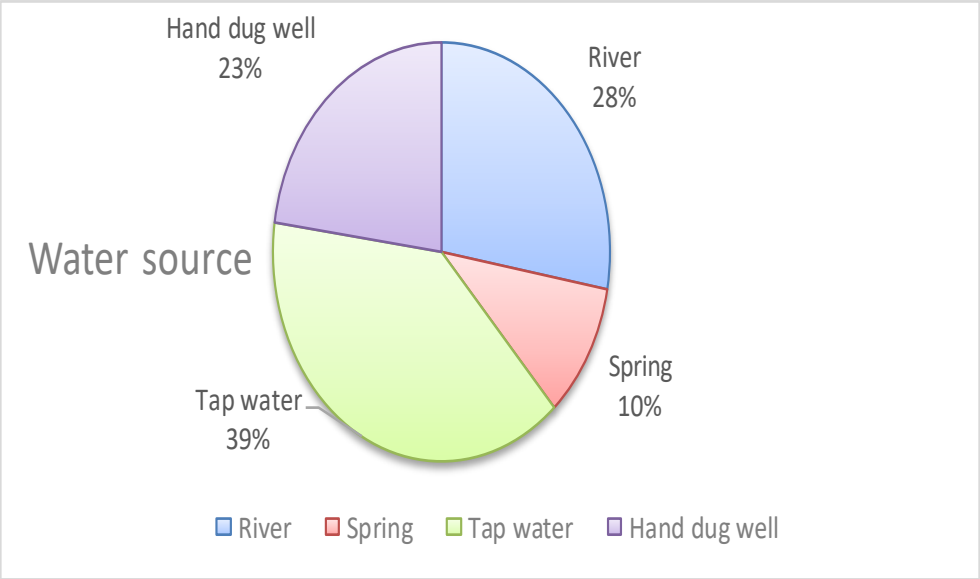


Figure 4.6: Main water sources for households in the study area.

Environmental and climate factors greatly affect the disseminated of CL. During the warmer months Sandflies are active in humid environments and at night, from dusk to dawn (Aklilu, E., Gebresilassie, A., Yared, S. et al. 2017). Therefore, living near watery areas may provide the necessary moisture needed for the sandfly.

4.2.2.1.5. Host related factors

Under this section, host factors which may contribute to the occurrence, transmission and prevention of the diseases are presented (Table 4.15). Keeping dry wood for future fire consumption for cooking is a custom in rural areas as electricity is unavailable. This may expose the household members to sandfly bite as it will create a perfect hiding place and resting place.

The majority (70.28%) households keep dry wood stored inside the house. Keeping woods near resident may serve as resting place for sandflies during the day times and increase the likelihood of bite. Travelling to an endemic area is one of the factors for acquiring the disease. However, the percentage of households with a history of travel was 12.39%.

Table 4.15: Description of host factors (n=1356).

Variables	Frequency (%)
Household waste disposal	
Closed	201(14.82)
Open	1,155(85.18)
Habit of sleeping at home yard/compound?	
No	896(66.08)
Yes	460(33.92)
Spending time near/at gorge early morning?	
No	972(71.68)
Yes	384(28.32)
Sit in home yard at down and evening?	
No	652(48.08)
Yes	704(51.92)
Sleep face and hand covered?	
No	297(21.90)
Yes	1,059(78.10)
Early morning work in the garden?	
No	898(66.22)
Yes	458(33.78)
Irrigation of crops at night?	
No	1263(93.21)
Yes	92(6.79)
Children play in forest at night.	
No	1,340(98.82)
Yes	16(1.18)

There is no vaccine or prophylactic treatment available therefore, the only way to avoid leishmaniasis is to prevent getting stung by a sandfly. The use of insecticide-treated bed nets was experienced by 40.86% of the individuals in the study. When asked about the sleeping condition, almost equal percentage of individuals said that they sleep on mat/matters with bed net (47.12%) and mat/mattress without a bed net (46.24%).

Sandflies feed on a variety of mammals, including wild animals, and the Leishmania parasite population in the forest spends its lifespan cycling between wild animals and vectors. Humans may become an unintentional host, infected by a vector, if they invade previously abandoned forests for farming or living. According to most of the participants (87.88%), the village is not expanding to a previously abandoned forest.

In the current study, of the 651 (48.0%) individuals who have a habit of sitting in the home yard at dawn/dusk, 25.34% do this always, 41.01% frequently and the rest 20.89% sometimes sit in the home yard at dawn/dusk. When the temperature is at a peak, 13.15% and 20.58% of the respondents in this study, prefer to work at night and in the early morning respectively and nearly half (47.01%) of them prefer working both at night and during the daytime. As explained earlier, working at night and early morning is one of the host factors determining the occurrence of leishmaniasis owing to the higher risk of the sandfly bite.

Majority of the respondents (63.13%) in this study do not take personal protective measures. The major types of personal protective measures practiced by the rest 36.87% are presented in Figure 4.7.

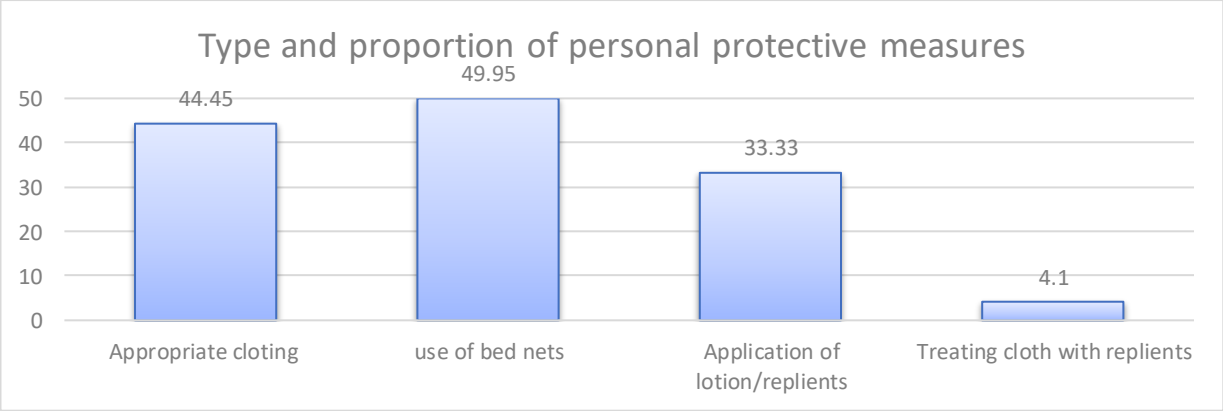


Figure 4.7: Type of personal protective measures used by the study population.

4.2.2.1.6. Animal factors

Different researchers show an association between CL and the presence of animals around the house. Therefore, in this section details of animal factors and their distribution among the households will be described. Hyraxes exist in working areas (47.20%), in villages (44.10%), and near the house (31.49%) and a half (50.07%) of the respondents verify the presence of hyrax burrow within 300-meter radius of the house. Figure 4.8 depicts the proportion of reservoir hosts (hyrax and bats) in the study area. When asked if they think the number of hyraxes or bats in the environment seems to be increasing, 39.09% confirmed that the reservoir host

population is indeed increasing time after time, and 15.34% claim that these hyraxes sometimes come into the household compound.

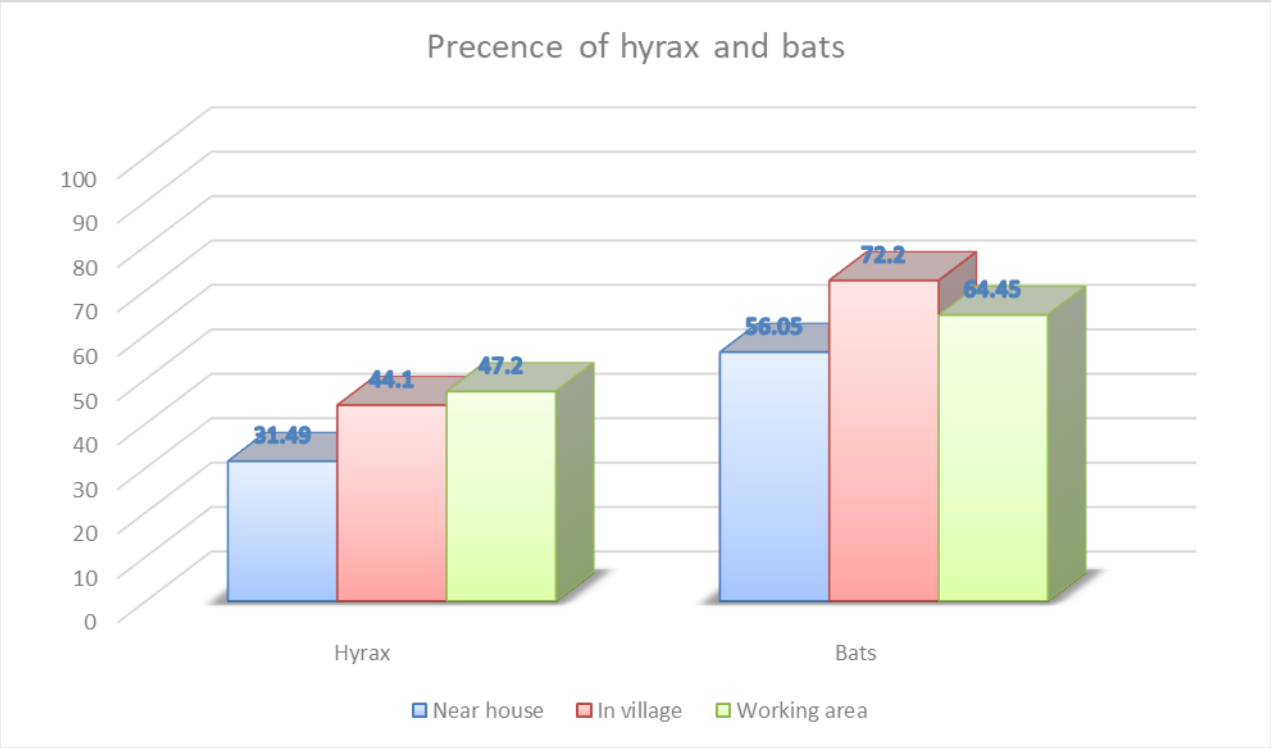


Figure 4.8: Presence of hyrax and bats in the vicinity of study participants.

The majority (80.53%) of the respondents in this study have domestic animals. Among these, cows (77.44%), donkeys (71.22), sheep/goat (40.09%), and poultry (47.92%) were mentioned. Most of the time these domestic animals are kept inside the household (29.3%) or in their stall near the household (61.44%). Four hundred and seven (30.01%) individuals said that there is a habit of dumping animal dung near the house. In the current study, pets (dog [in 64.99%] and cat [52.03%]) are present in most of the households (77.58%).

4.2.2.2. Analysis of the association of factors with disease occurrence

The epidemiology of CL can be influenced by a favourable environment for vectors, intensive agricultural practices, the presence of a reservoir, and migration. All of these variables are present in Ethiopia's northern and southern regions (Assefa

2018:17). To identify factors that are associated with leishmaniasis, selected variables from demographic characteristics, household factors, environmental factors, host factors, and animal factors were used. A detailed description of the variables in each category and the type of statistical methods used.

4.2.2.2.1. Demographic characteristics and residential futures with cutaneous leishmaniasis

Following the two methods described in this chapter under the data management section, the variable religion was excluded because it shows no variability within the categories as nearly all respondent belongs to one category (Orthodox religion). Two categories (higher education with a frequency of five and secondary level education with a frequency of 269) in level educational were merged to make a new variable category called "secondary and above". This was done owing to the small number of observations with higher education category. Variable measuring, if the individual was born and raised in the same district, was not included in the analysis because it shows no variability (105 for coming from other district and raised in the district against 1252 who are born and raised there). Measurements of types of occupations other than farming were excluded because of small observations and no variability among categories. Therefore, six variables (kebele, age, sex, education, occupation related to farm and marital status) describing demographic characteristics of the respondents were selected.

The dependent variable was the presence of infection and for univariate logistic regression each independent variable was treated with this variable separately. In multivariate logistic regression, the six independent variables selected above were considered in the initial model, and then stepwise backward selection method was used to identify the most appropriate variables to be included in the model. Regarding the bivariate and multivariate logistic regression of demographic variables against the presence of disease in the person, only a residential kebele was significant in both cases (Table 4.16). Accordingly, in bivariate logistic regression Beki and Kola nurena kebeles were significantly linked with the disease. However, in multivariate logistic regression, individuals from Kola Nurena kebele

have 6.85 times higher odds of having the disease compared to individuals from Semero [AOR=6.85; 95% CI, 3.860-1.220e+01, p-value <0.0001].

Table 4.16: Demographic factors associated with cutaneous leishmaniasis.

Factor	Bivariate	
	p-value	**COR (95%CI)**
Kebele		
Semero		1
Beki	0.011 *	2.56(1.184-5.179)
Genete Mariam	0.2943	0.72(0.381- 1.307)
Kela Zuria	0.5112	0.77(0.343- 1.586)
Kola Nurena	<0.0001 ***	7.42(4.316- 12.81)
Michael	0.1004	1.63(0.893- 2.923)
Gender		
Female		1
Male	0.576	0.89(0.619-1.303)
Eduction		
Able to read and write		1
Illiterate	0.8638	0.05(0.555-1.689)
Informal education	0.3998	0.88(0.022-2.153)
Primary	0.4865	0.20(0.466-1.469)
Secondary & above	0.0789	0.60(0.275-1.071)
Marital status		
Married		1
Not married	0.613	0.90(0.614-1.322)
Farm related work		
No		1
Yes	0.357	0.174(0.820-1.726)

4.2.2.2.2. Association between household factors and cutaneous leishmaniasis

In the data selection procedure, wall type and presence of functional latrine were removed from logistic regression due to lack of variability among categories. Data measuring wall condition were subjected to change to make the variable sound for the identification of its effect on the disease. Therefore, the previous four categories (cracked/holes formed, holes formed, cracked, and no crack/holes) were merged into categories which are no crack/holes formed and cracked/hole formed/both (which contained all values of the first three categories previously mentioned).

Then five household factors (wall condition, type of floor, type of roof, latrine condition, and presence of screen on window or door) were selected for bivariate and multivariate logistic regression tests. As a result, the presence of a screen on the window or the door was significantly related to disease occurrence in both tests (Table 4.17). Houses with no screen are twice at risk of getting infected with leishmaniasis [AOR=2.49; 95% CI, 1.448-4.571, P-value 0.001].

Table 4.17: Association of household factors with cutaneous leishmaniasis.

Factor	**Uivariate**	
	p-value	**COR (95%CI)**
Screen on window/door		
Yes		1
No	0.0121*	2.00(1.198-3.565)
Floor type		
Cemented		1
Cow dung plastered	0.4593	2.14(0.443-38.429)
Mud	0.1328	4.88(0.924 - 90.379)
Roof type		
Dry grass		1
stone/made of fabric	0.828	0.79(0.043-4.178)
steal	0.820	1.04(0.701-1.599)
Latrine condition		
Good		1
Absent	0.494	1.34(0.543-3.027)
Bad	0.872	0.94(0.486-1.813)
Fair	0.755	1.08(0.668-1.809)

4.2.2.2.3. Environmental factors and cutaneous leishmaniasis

Environmental factors selected to be affecting CL presence are the existence of farm in 300m radius of the house, presence of acacia and sensel trees, and presence of cave or gorge near home, near the village and working area. The result of the bivariate and multivariate analysis shows a significant relationship between the presence sensel tree and cave or gorge near home with disease occurrence (Table 4.18). This result shows that individuals living near sensel tree are at double risk of acquiring the disease [AOR=2.31; 95% CI, 1.507-3.555, P-value 0.0001]. Moreover, the presence of a cave or gorge near home increases the likelihood of infection by seven-fold [AOR=7.02; 95% CI, 4.419-11.273, p-value <0.0001].

4.18: Environmental factors and cutaneous leishmaniasis.

Factor	Univariate	
	p-value	**COR (95%CI)**
Presence of Sensel tree		
No		1
Yes	0.001**	1.89(1.264-2.815)
Cave/gorge near home		
No		1
Yes	<0.0001 ***	4.42(3.023-6.478)
Farm with in 300 radius		
No		1
Yes	0.225	0.78(0.526-1.175)
Accacia tree near home		
No		1
Yes	0.981	0.99(0.686-1.445)
Cave/gorge near village		
No		1
Yes	0.24	1.25(0.862-1.816)
Cave/gorge near workplace		
No		1
Yes	0.305	1.22(0.833-1.766)

‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05

4.2.2.2.4. Host related factors and their relation to cutaneous leishmaniasis

Selected host-related variables contributing to CL occurrence were twelve. These include travel history in the family; sleeping at home yard/compound; use of impregnated bed nets; working in the farm garden early in the morning; spending time near the cave/gorge; sitting at the home yard at the evening; frequency of sitting at the home yard; sleeping hand and face covered; irrigation crops at night; use of personal protective measures; sleeping condition in the house and keeping dry woods stored inside the house. Out of the twelve of these variables, nine of these show a significant relationship with the occurrence of the diseases in bivariate logistic regression analysis (Table 4.19). These variables are travel history, sleeping in home yare/compound, use of insecticide-treated bed nets, working in the farm early morning, spending time near gorge/cave, siting in the compound at night, sleep and hand face covered, irrigation at night and personal protective measures.

Table 4.19: Host factors affecting transmission of CL.

Factor	Univariate	
	p-value	COR (95%CI)
Travel history		
Yes		1
No	0.002**	6.17(2.299-25.280)
Sleep home yard/compound		
No		1
Yes	<0.0001 ***	5.34(3.592-8.078)
Use of impregnated bed nets		
Yes		1
No	<0.0001 ***	2.79(1.814-4.453)
Working at farm early morning		
No		1
Yes	0.0228 *	1.55(1.059- 2.250)
Spending time near gorge/cave		
No		1
Yes	<0.0001 ***	2.22(1.520-3.237)
Siting in compound at night		
No		1
Yes	<0.0001 ***	2.22(1.500- 3.337)
Sleep hand/face covered		
Yes		1
No	<0.0001 ***	4.16(2.838- 6.088)
Irrigation at night		
No		1
Yes	4.91e-16 ***	7.16(4.423-11.486)
Use of personal protective measures		
Yes		1
No	<0.0001 ***	6.69(3.730- 13.303)
Keep dry woods		
No		1
Yes	0.261	0.79(0.542-1.190)

‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05

Man-made factors are mainly responsible for worldwide increase of leishmaniasis which grant breeding and resting sites of the vector (WHO 2020). Man-made environmental management such as the building of a bridge in "Silti" town in Southern Ethiopia, on reverse, has resulted in the CL epidemic owing to the creation of an ideal habitat for sandflies and hyraxes to replicate under it (Lemma 2018:317).

4.20: Multivariate logistic regression of host factors.

Factor	Multivariate logistic regression	
	p-value	AOR (95%CI)
Travel history past year		
Yes		1
No	0.00256 **	6.87(2.274-30.163)
Sleep home yard/compound		
No		1
Yes	<0.0001 ***	12.91(4.565-36.868)
Spending time near gorge/cave		
No		1
Yes	0.01745 *	2.29(1.161-4.569)
Sleep hand/face covered		
Yes		1
No	<0.0001 ***	8.68(4.943-15.643)
Irrigation at night		
No		1
Yes	<0.0001 ***	4.14(2.209-7.798)
Use of personal protective measures		
Yes		1
No	0.04075 *	2.23(1.062-5.001)

'***' 0.001 '**' 0.01 '*' 0.05

In multivariate logistic regression factors such as having no history of travel [AOR=6.87; 95% CI, 2.274-30.163], sleeping in home yard/compound [AOR=12.91; 95% CI, 4.565-36.868], spending time near gorge/cave [AOR=2.29; 95% CI,1.161-4.569], not covering hand/face while sleeping [AOR=8.68; 95% CI, 4.943-15.643], irrigation at night [AOR=4.14; 95% CI, 2.209-7.798] and not using personal protective measures [AOR=2.23; 95% CI, 1.062-5.001] were all significantly related with CL as shown in Table 4.20. Accordingly, those individuals with travel history have 7 times higher risk of infection. Sleeping in the home yard/compound, spending time near the gorge/cave, not covering hand/face while sleeping, irrigation at night, and not using personal protective measures increase the likelihood of being infected by 12.9, 2.8, 8.6, and 4.1-fold, respectively.

4.2.2.2.5. Animal-related factors affecting cutaneous leishmaniasis occurrence

Animal factors selected for logistic regression were the existence of hyrax and bats near the house, near the working area and near the village, dumping animal dung

near the house, presence of domestic animals, and animal burrow within 300 metres radius of the house.

As presented in Table 4.21, individuals with hyrax around the house [AOR=2.49; 95% CI, 1.679-3.708, p-value <0.0001] and have a habit of dumping cow dung near the house [AOR=2.35; 95% CI, 1.549- 3.557, p-value <0.0001] are 2.4 and 2.3 times get CL respectively, than those who live far from hyraxes and do not dump cow dung near their house.

Table 4.21: Association of animal factors to CL.

Factor	**Univariate**	
	p-value	**COR (95%CI)**
Hyrax around house		
No		1
Yes	<0.0001 ***	2.33(1.602-3.390)
Hyrax at working area		
No		1
Yes	0.0391 *	1.48(1.021-2.159)
Hyrax at village		
No		1
Yes	0.274	1.23(0.847-1.783)
Dumping cow dung near house		
No		1
Yes	0.00401 **	1.74(1.189-2.544)

‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05

4.2.2.2.6. Association analysis of all factors with disease occurrence

All variables include the following:

- Demographic characteristics (kebele, age, sex, education, occupation related to farm and marital status).
- Household factors (wall condition, type of floor, type of roof, condition of latrine and presence of screen on window/door).
- Environmental factors (presence of farm in 300 radius, presence of acacia, presence of sensel, presence of cave/gorge near home near village and near working area).

- Host factors (travel history in the family, sleeping at home yard/compound, use of impregnated bed nets, working in the farm garden early in the morning, spending time near the cave/gorge, sitting at the home yard at the evening, frequency of sitting at home yard, sleeping hand and face covered, irrigation crops at night, use of personal protective measures, sleeping condition in the house and keeping dry woods stored inside the house) and
- Animal factor (existence of hyrax and bats near house, near working area and near village, dumping animal dung near house, presence of domestic animals and animal burrow with on 300 radius of the house).

These variables were chosen based on the criteria established in the preceding logistic analysis and combined to perform stepwise logistic regression using all variables in order to determine the best predictor(s) for the absence and presence of CL in the study area throughout the study period.

As a result, kebele (Kola Nurena) [AOR=2.88; 95% CI, 1.037- 8.005], sleeping at home yard [AOR=30.55; 95% CI, 11.288-88.450], irrigation at night [AOR=6.80; 95% CI, 2.683-18.214], presence of cave/gorge near home [AOR=1.66; 95% CI, 0.953-2.919], spending time near gorge early morning [AOR=3.90; 95% CI, 1.589-9.782] and presence of hyrax near house [AOR=6.94; 95% CI, 3.092-16.513] were exposing factors to the disease. Individuals living in Kola Nurena Kebele are 2.8 times more likely to get infected and individuals who have a habit of sleeping at home yard, irrigating crops at night and spending time near cave/gorge early morning are at a higher risk to CL 30.6, 6.8 and 3.9 times, respectively. The presence of hyrax and cave/gorge near home are found to expose individuals to the disease 6.9 and 1.6 times than those who live far from hyrax colony and cave/gorge.

Table 4.22: Analysis of all risk factors with the disease.

Factor	Multivariate logistic regression	
	p-value	AOR (95%CI)
Kebele		
Semero		1
Beki	0.4862	1.68(0.375-7.205)
Genete Mariam	0.0024**	0.16(0.046-0.506)
Kela Zuria	<0.0001 ***	0.013(0.003- 0.049)
Kola Nurena	0.0411*	2.885(1.037- 8.005)
Michael Semero	<0.0001 ***	0.066(0.020-0.203)
Screen window/door		
No		1
Yes	<0.0001 ***	0.088(0.035-0.214)
Cave/gorge near home		
No		1
Yes	0.0729.	1.665(0.953-2.919)
Hyrax near home		
No		1
Yes	<0.0001 ***	6.943(3.092-16.513)
Sleep home yard/compound		
No		1
Yes	<0.0001 ***	30.558(11.288-88.450)
Spending time near gorge/cave		
No		1
Yes	0.003199 **	3.906(1.589-9.782)
Sleep hand/face covered		
No		1
Yes	<0.0001 ***	0.056(0.024-0.124)
Irrigation at night		
No		1
Yes	<0.0001 ***	6.805(2.683-18.214)
Use of personal protective measures		
No		1
Yes	<0.0001 ***	0.143(0.051- 0.362)

On the other hand, living in Kela Zuria kebele [AOR=0.01; 95% CI, 0.003- 0.049], Michael Semero [AOR=0.07; 95% CI, 0.020-0.203] and Genete Mariam [AOR=0.16; 95% CI, 0.046-0.506], use of personal protective measure [AOR=0.14; 95% CI, 0.051- 0.362], sleeping hand/face covered [AOR=0.05; 95% CI, 0.024-0.124] and presence of screen on window door [AOR=0.088; 95% CI, 0.035-0.214] were protective factors (Table 4.22). Living in Kela Zuria, Micheal Semero and Genete Mariam reduce the log odds of infection by 4.2, 2.7 and 1.8, respectively. In

addition, the use of personal protective measure, sleeping hand/face covered and having screen on window/door reduce the log odds of CL by 1.9, 2.8 and 2.4.

Most of the factors affecting disease occurrence in this communities are host/behavioural factors (use of personal protective measures, irrigation at night, sleep hand/face covered, putting screen on window and door, sleeping at home yard and spending time near gorge/cave) that can be changed through effective awareness creation campaigns.

4.2.2.3. Multiple correspondence analysis of selected factors

In a large and complex dataset, an Exploratory Data Analysis (EDA) technique such as Multiple Correspondence Analysis (MCA) can be used to to identify key factors contributing to CL. Informations can be extracted from datasets and changed to a more understandable sturcure trough EDA. MCA is a powerful technique with more conventional graphical presentation and easier interpretations than log-liner models (Jalayer & Zhou 2016).

The researcher considers all twelve variables for multiple correspondence analysis which show significant statistical association with the disease in multiple logistic regression of demographic, household, environmental, host and animal factors. These variables were dumping animal dung near house and presence of hyrax near house (as animal factor), presence of sensel tree and cave gorge near home (as environmental factors), presence of screen on window or door of the house (as household factor), residential kebele (demographic factor) and use of personal protective measures, irrigation at night, sleeping covering hand and face, spending time near gorge/cave, sleeping at home yard/in compound and travel history (as host factors). The variables explaining demographic characteristics (age, sex, kebele, marital status, education, and occupation) of the study participants were also included in the MCA as a supplementary variable.

To pinpoint the main influencing factors, R statistical software (Version 3.02) and a package called FactoMineR were used. Graphical views in MCA aid understanding

of relationships of variables. If variables share certain similarities, then they will be presented closer to each other in the two-dimensional graph of the data (Das & Sun, 2014:9). The magnitude of information associated with each dimension is called eigenvalue which has a value ranging 0-1. The eigenvalue shows the total variance between variables (Fritzell, Raude, Addel, et al. 2016:6).

The first and second dimensions have larger eigenvalues than the other dimensions, according to the MCA analysis performed in this section, and no outliers were found. The first two dimensions show strong relationships between variables.

The percentage variability explained by dimension one is 23.57% and by dimension two is 15.93%. This suggests that the first two dimensions of the analyses (One and Two) express 39.5% of the total dataset inertia, suggesting that the plane describes 39.5% of the people (or variables) cloud overall variability. This figure is higher than the standard value of 23.14%, indicating that the plane's variability is significant. The 0.95-quantile of the inertia percentages distribution found by simulating 2080 data tables of comparable size based on a uniform distribution is used as the reference value. (Fritzell, Raude, Addel, et al. 2016:6) (Figure 4.9).

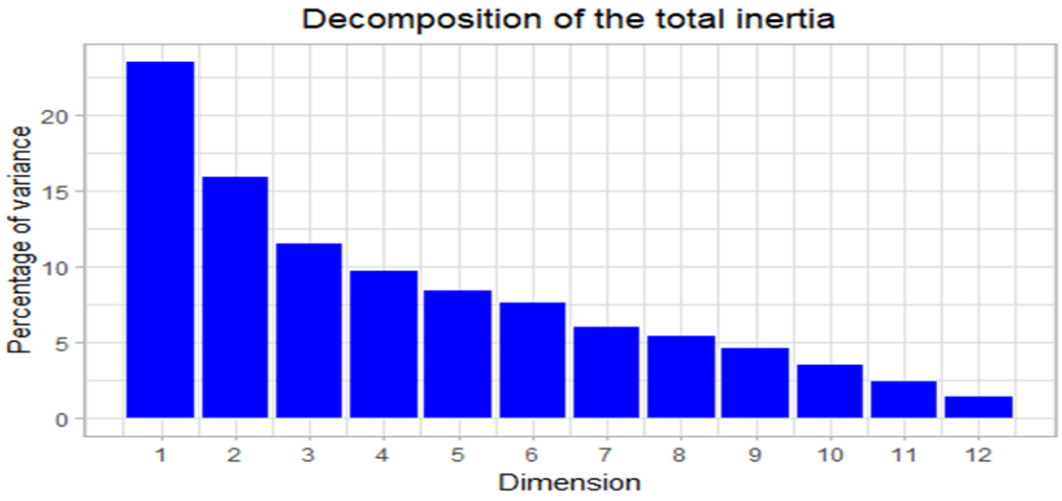


Figure 4.9: Decomposition of the total inertia.

For understanding of relationships between variables, the first two dimensions are

used. From Figure 4.9 above, it can be observed that consideration of the third dimension might be interesting since high percentage of inertia (variablility) is expressed by it.

Figure 4.10 shows relationship between variables and variables more closely associated are placed together. As a result, disease status (*CL_present*) is correlated to habit of spending time near the gorge early morning (*spending near at gorge early morning*), residential kebele, presence/absence of screen on window and doors (*screen window/door*), presence/absence of cave or gorge near home (*cave/gorge home*) and irrigation of crops at night (*irrigation crops at night*).

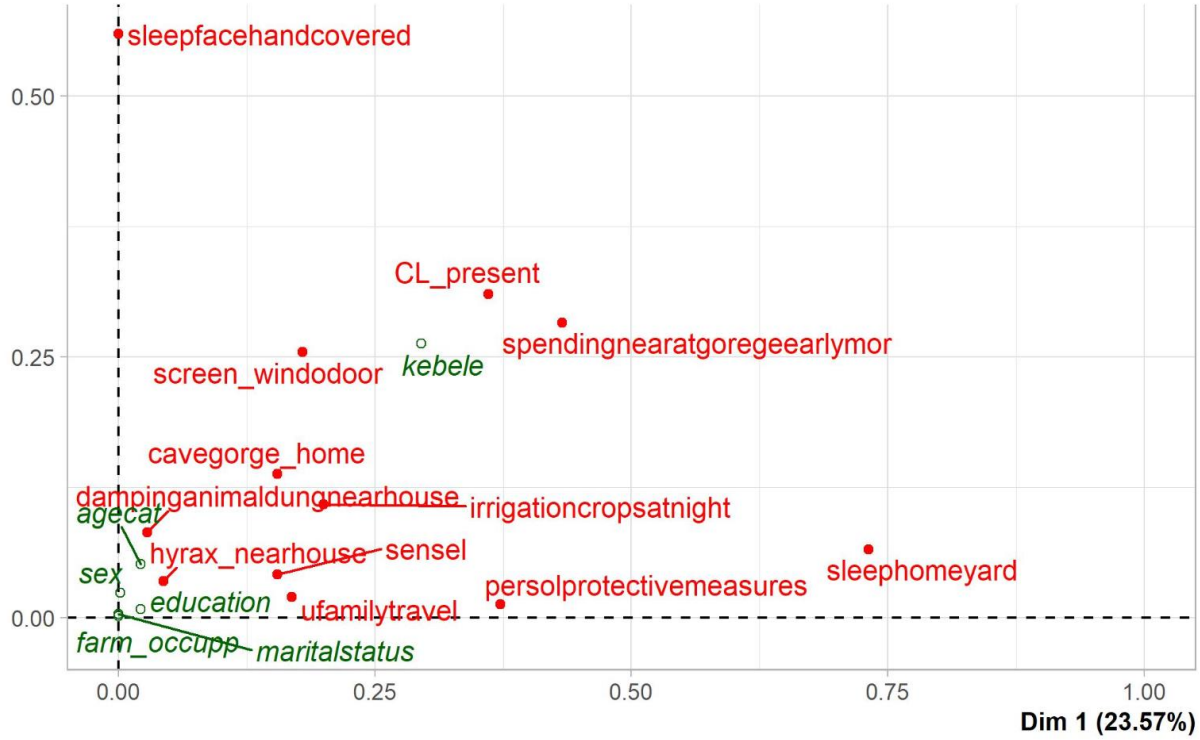


Figure 4.10: Description of variable representation.

MCA can also be used to see associations between variable categories. Looking into the variable categories displayed in the Figure 4.11, the presence of CL is associated with those individuals who irrigate crops at night, live in Kola nurena and Beki kebele, who are less than 10 years old, and those who sleep without covering their face and hand. In addition, households with positive leishmaniasis cases are

those which damp animal dung near house, and those that have cave/gorge, sensel tree and hyrax nearby the house. Families with no history of travel are also associated with presence of the disease.

The absence of CL is seen associated with sleeping hand face covered, absence of cave/gorge near home, no history of travel, no habit of dumping animal dung near home and absence of hyrax near home. Figure 4:12 depicts the same MCA map but without the supplementary variables to simplify the map and understand relationships.

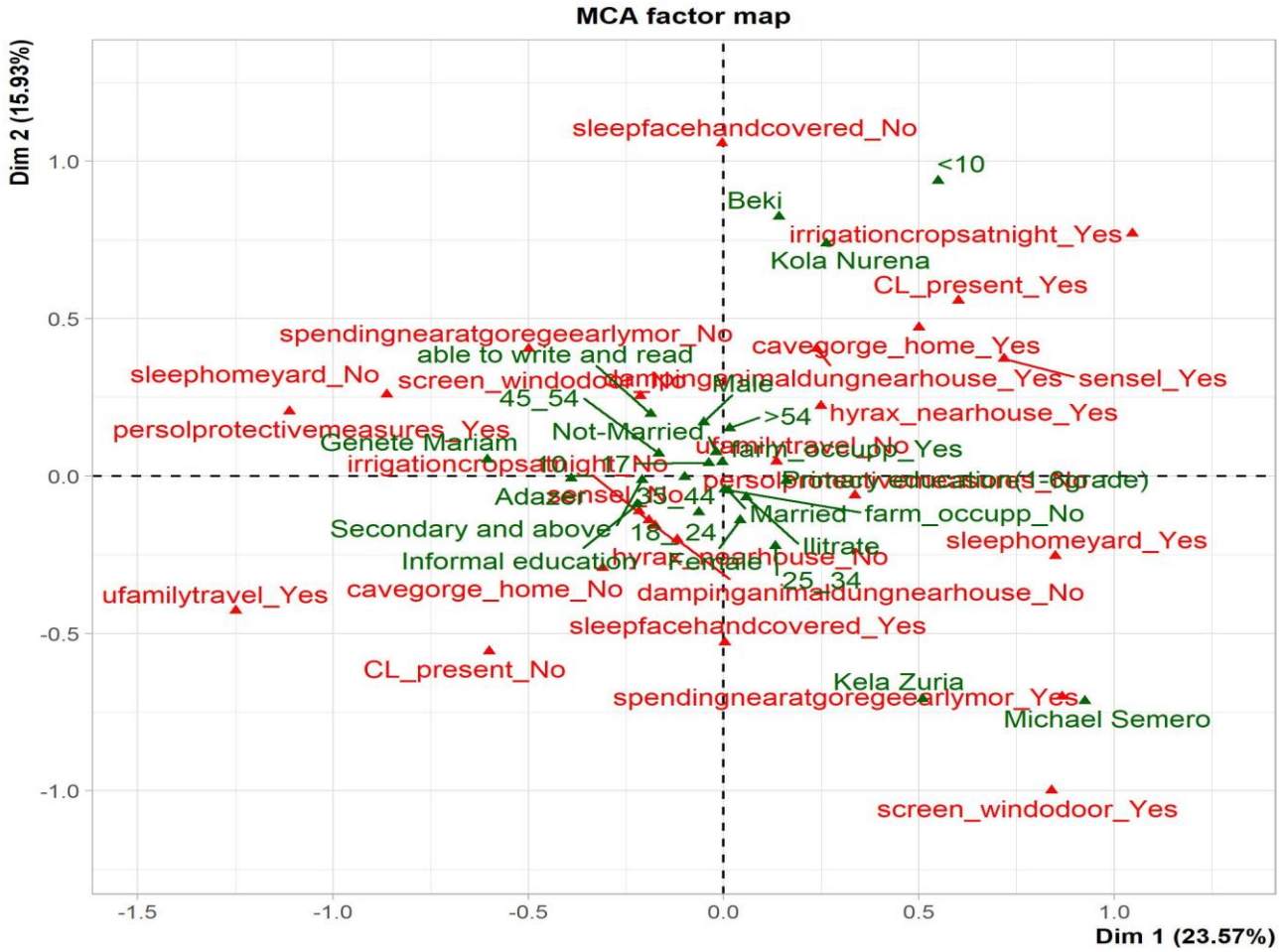


Figure 4.11: MCA factor map (with supplementary variables).

This figure shows distribution of variables (non supplementary variables in Red and supplementary variables in Green) in the map.

Figure 4.12: MCA factor map (without the supplementary variables).

Looking into the distribution of individuals on the map (Figure 4.13) below, the distribution of study participants (Roman numbers) can be viewed according to their disease status and their associations with factors of the disease.

In dimension one, it can be seen that:

The group (B) in which the individuals *223*, *162* and *194* stands (characterised by a positive coordinate on the axis) is sharing:

- high frequency for factors like *CL_present_(Yes)*, *cave/gorge home (Yes)*, *sleep face hand covered (No)*, *irrigation crops at night (Yes)*, *hyrax near house (Yes)*, *sensel (Yes)*, *Kola Nurena*, *age category =<10*, *spending near gorge early morning (No)* and *dumping animal dung near house (Yes)* (factors are sorted from the most common).

- low frequency for factors like *are you infected (No)*, *cave/gorge near home (No)*, *sleep face hand covered (Yes)*, *irrigation crops at night (No)*, *hyrax near house (No)*, *sensel (No)*, *spending near gorge early morning (Yes)*, *kebele=Kela Zuria*, *dumping animal dung near house (No)* and *you or family travel (Yes)* (factors are sorted from the rarest).

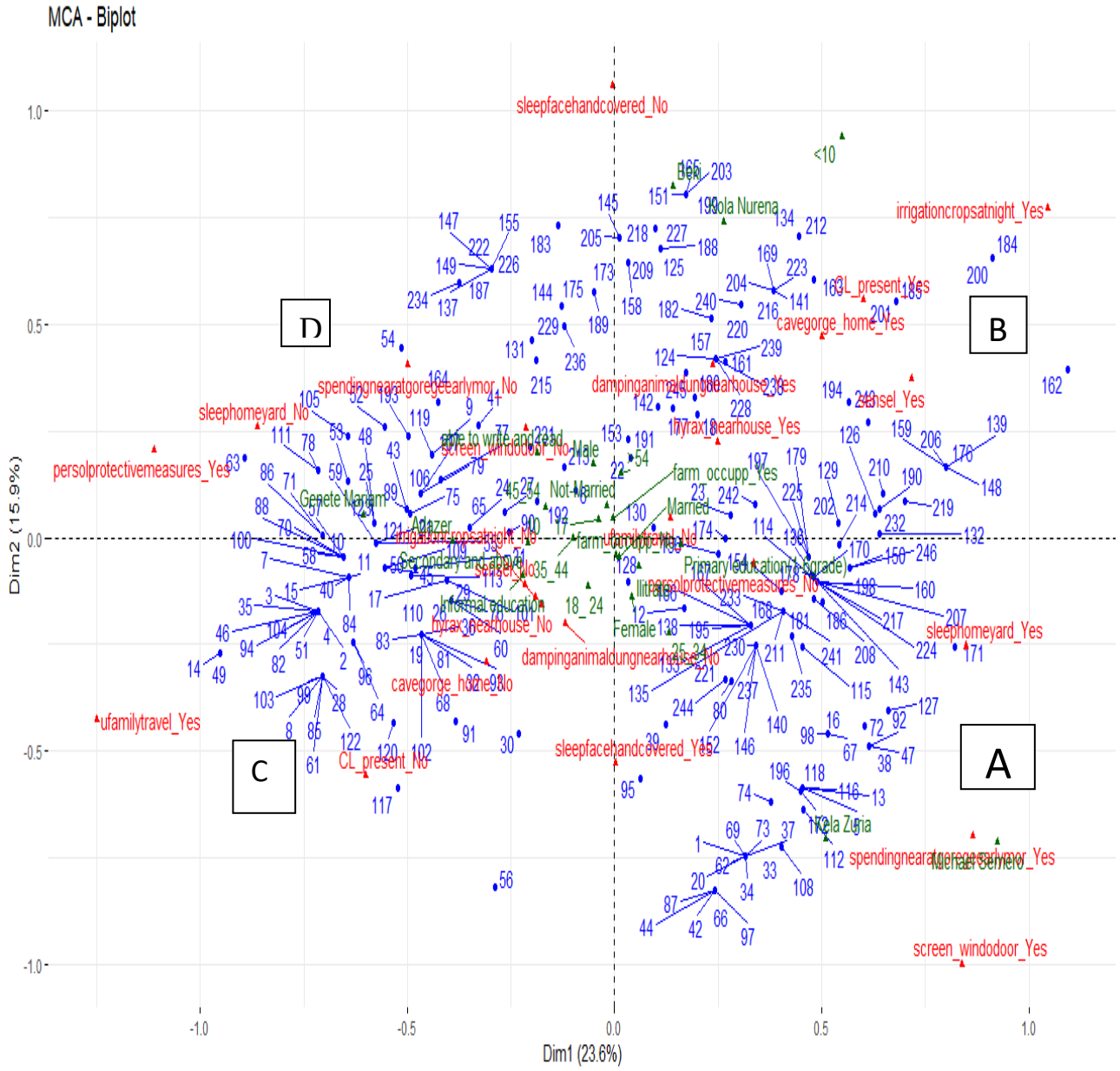


Figure 4.13: MCA factor map (with the distribution of individuals).

In this figure association of main variables (in Red) and supplimentry variables (in Green) with study individuals (in Blue) is illustrated.

Dimension 2 opposes individuals such as *117*, *40*, and *83* which are on the top of the graph to individuals on the bottom of the graph both of which are illustrated by a strongly positive coordinate on the axis.

The group () (characterised by a negative coordinate on the axis) is sharing:

- high frequency for factors like *sleep home yard (No)*, *are you infected (No)*, *cave/gorge near home (No)*, *spending near gorge early morning (No)*, *hyrax near house (No)*, *personal protective measures (Yes)*, *screen window/door (No)*, *sensel (No)*, *kebele=Adazer*, and *sleep face hand covered (Yes)* (factors are sorted from the most common).
- low frequency for factors like *sleep home yard (Yes)*, *are you infected (Yes)*, *cave/gorge home (Yes)*, *spending near gorge early morning (Yes)*, *hyrax near house=hyrax near house (Yes)*, *personal protective measures (No)*, *screen window/door (Yes)*, *sensel (Yes)*, *kebele=Michael Semero*, and *sleep face hand covered (No)* (factors are sorted from the rarest).

From the individual MCA (map showing the distribution of individuals), those individuals with the disease and those without CL have distinctive characteristics (demographic, household, environmental, host, and animal factors) which separate them into two clouds (Figure 4.14). This means there are certain characteristics that expose an individual to CL and there are some factors that protect another from the disease. "Yes" on the map means the presence of CL and "No" shows a cloud of individuals with no CL.

Knowing that factors can define the disease status of the community is particularly important for informing the design of prevention and control strategies specific to the community. For health decision-making and planning processes, correct information on disease burden and risk factors are vital (WHO 2020). From the individual MCA, clouds indicate variability among two groups of individuals (with and without CL) based on different factors. Therefore, interventions based on the alteration of factors can be effective to change disease transmission within the community.

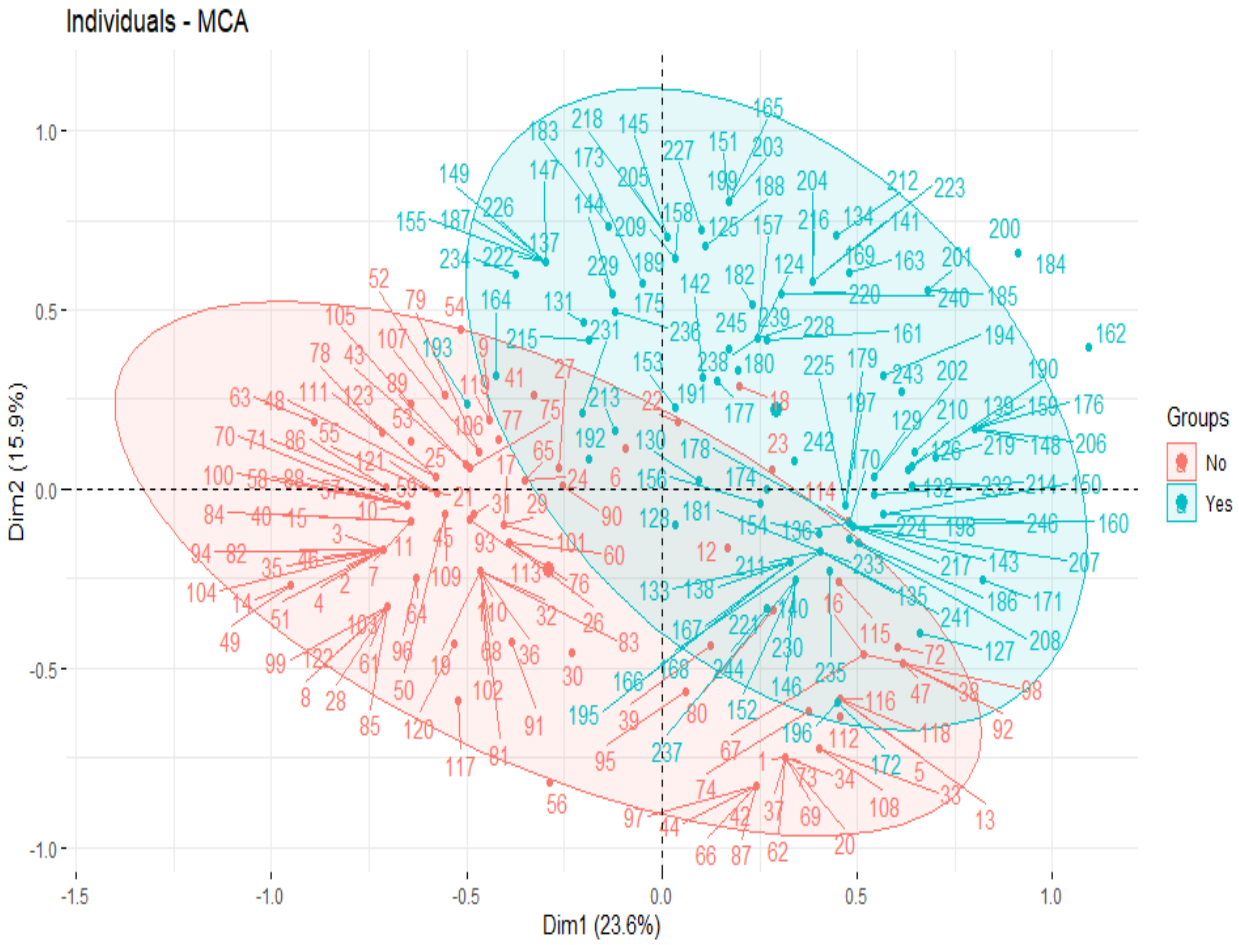


Figure 4.14: Multiple correspondence analysis factor map of individuals.

In this figure, individuals (those who have CL in Green and those with out CL in pink) are shown to have distinict groups based on their CL status.

After looking into the distinct differentiation between individuals that are with CL compared to those that are without CL, another MCA map was produced by selecting exposing factors categories. These include not covering hand and face when sleep, irrigation crops at night, a habit of dumping animal dung near the house, presence of cave/gorge near home, presence of sensel tree and hyrax near home, use of protective measures, having a habit of sleeping at the home yard and positive history of travel) and disease status (Yes and No) (Figure 4.15). This variable category MCA shows that except a positive history of travel, all other factors have an association with disease occurrence and of these factors, positive answer for the presence of cave/gorge, sensel tree, hyrax near home and habit of

irrigation at night, dumping animal dung near home and using protective measures closely associated with CL infection. Therefore, future intervention strategies can be designed prioritising these factors for consideration to prevent and control. In addition to the creation of a feasible prevention strategy (specific to factors affecting this community), this approach can also impose proper utilisation of limited resources and budget thus providing more effective and less costly.

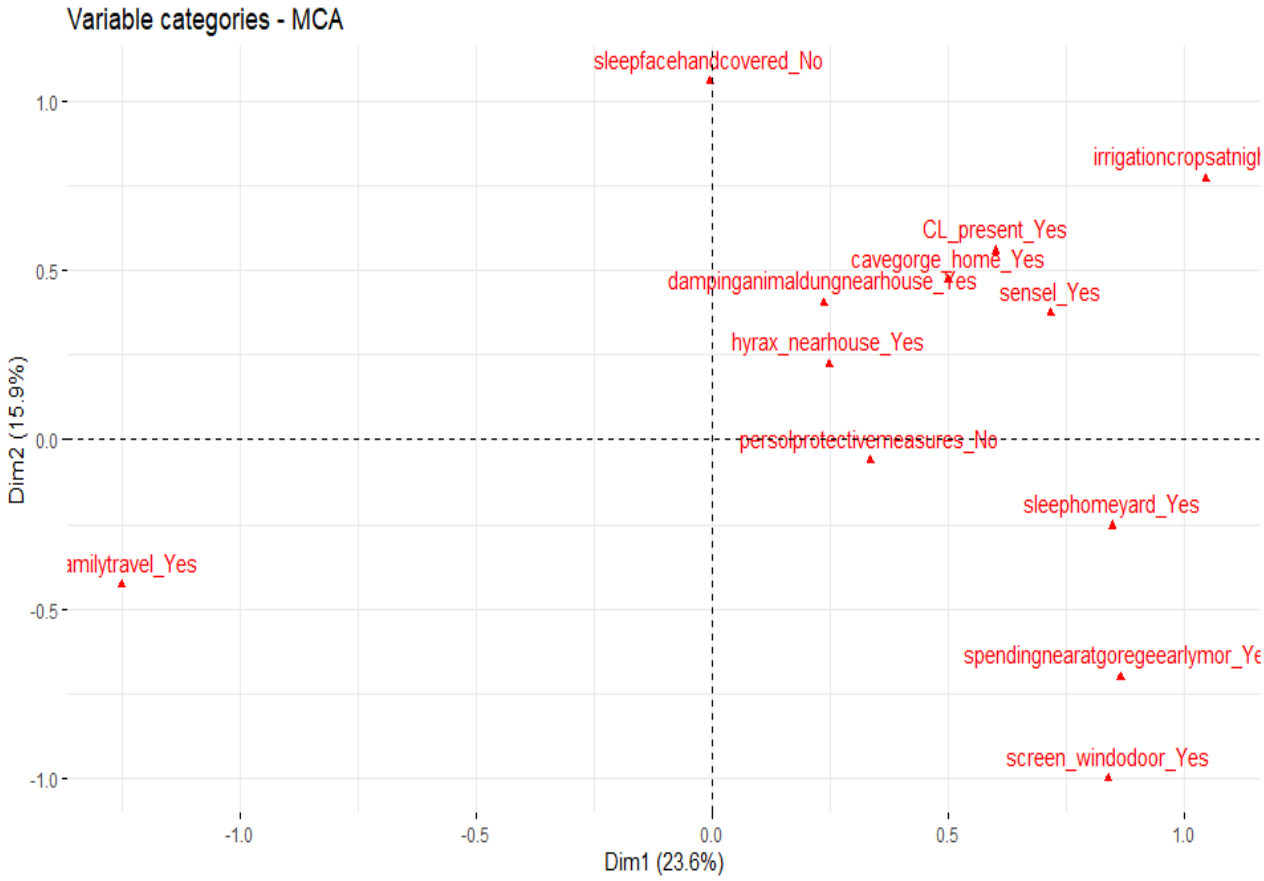


Figure 4.15. Variable MCA with exposing factors.

4.2.2.4. Hierarchical cluster analysis

After the MCA, a hierarchical cluster analysis was done to establish the natural groups of observations regarding CL status. Data points that have similar characters can be grouped via hierarchical clustering. These groups are termed as clusters, and because of hierarchical clustering, we get a set of clusters where these clusters are different from each other. Clustering help divide the study

population into similar groups therefore groups are separated by sharing similar traits and are assigned into clusters (Zolfaghari, Khosravi, Shahriyari, Jabbari & Abolhasani 2019:4). There are five types of clustering methods, comprising hierarchical, partitioning, density-based, fuzzy and model-based clustering (Hamid, Meaney, Crowcroft, Granerod & Beyene 2010). Hierarchical clustering is a method for spotting groups in the dataset. The outcome of hierarchical clustering is called a a dendrograma which is a tree-based image of the objects. To subdivide groups the dendrogram is cut at a pre-setted similarity level (Bock 2020).

Ward's minimum-variance hierarchical clustering approach with standardisation of integrated variables was performed in the R software package in this section, as stated by Fritzell (2016). Individuals with the shortest distance apart were hierarchically connected first, using the hierarchical agglomerative approach. Depending on the type of link function, different hierarchical techniques are available (Zolfaghari, Khosravi, Shahriyari, Jabbari & Abolhasani 2019:4). Using the complete linkage or furthest neighbor approach, the distance between two clusters was estimated as the maximum of individual-to-individual distances (Zolfaghari, Khosravi, Shahriyari, Jabbari & Abolhasani 2019:4).

A dendrogram is a branching diagram that characterizes the relationships of similarity among groups and the main use of a dendrogram is define the best way to assign objects to clusters (Bock 2020).

Looking at the below dendrogram, two clusters are seen as two branches that occur at about the same horizontal distance. Figure 4.16 above is also extended information to the fact that the study individuals can be clustered into two distinct groups. As Cluster 1 (in black) and cluster 2 (in red).

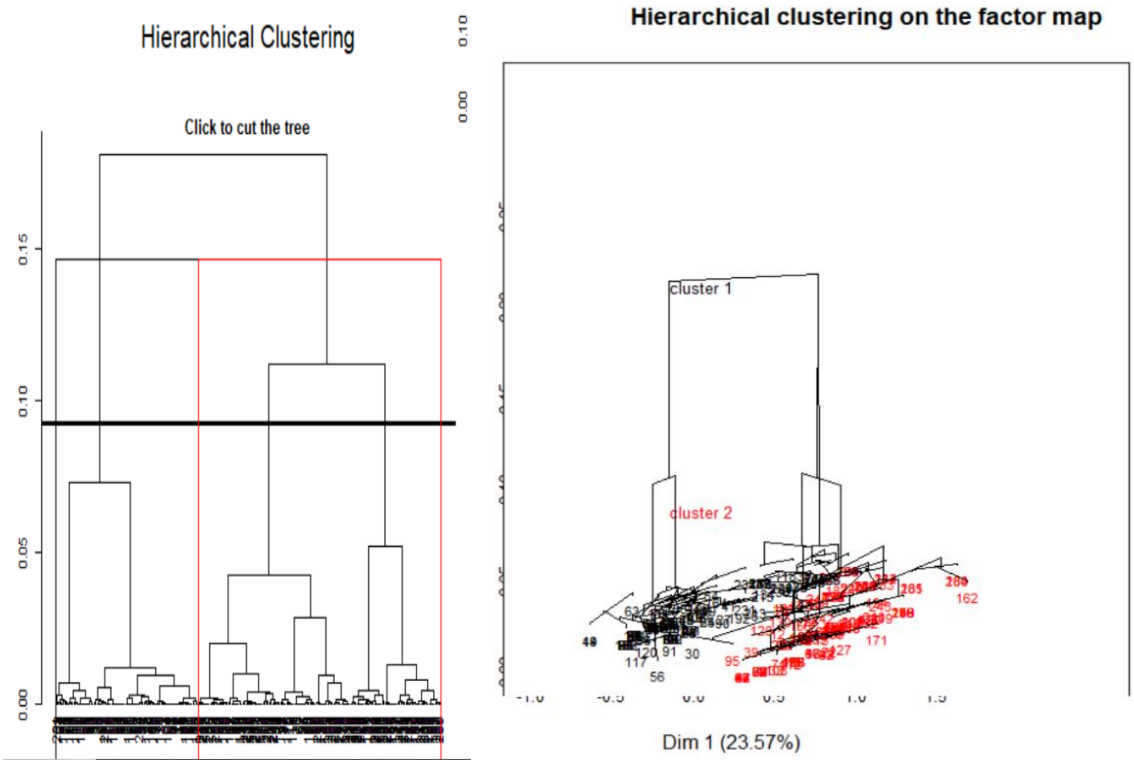


Figure 4.16: Dendrogram generated by the hierarchical clustering (left) and a three-dimensional plot combining the hierarchical clustering and the factorial map (right).

Figure 4.17 shows the visualisation of individuals on the principal component map, as well as the color of individuals based on which cluster they belong to. The results of the cluster analysis show that there are two distinct groups of individuals sharing different characteristics. Individuals in cluster 1 are without disease infection, do not spend time at the gorge early morning, use personal protective measures, and do not irrigate crops at night. In contrast, participants in cluster two are characterised by being clinically positive to CL, having a habit of sleeping at the home yard, spending time at the gorge early morning, and irrigating crops at night. Individuals in these groups also do not use personal protective measures against CL.

In line with the findings of the multiple correspondence analysis, the hierarchal cluster analysis also presents proof of the role played by different factors for the occurrence of the disease. Coupled with the result of the logistic regression, it can

be assumed that identified demographic, household, environmental, host, and animal factors are not only significantly associated with CL but also can group individuals in the study population into two distinct groups.

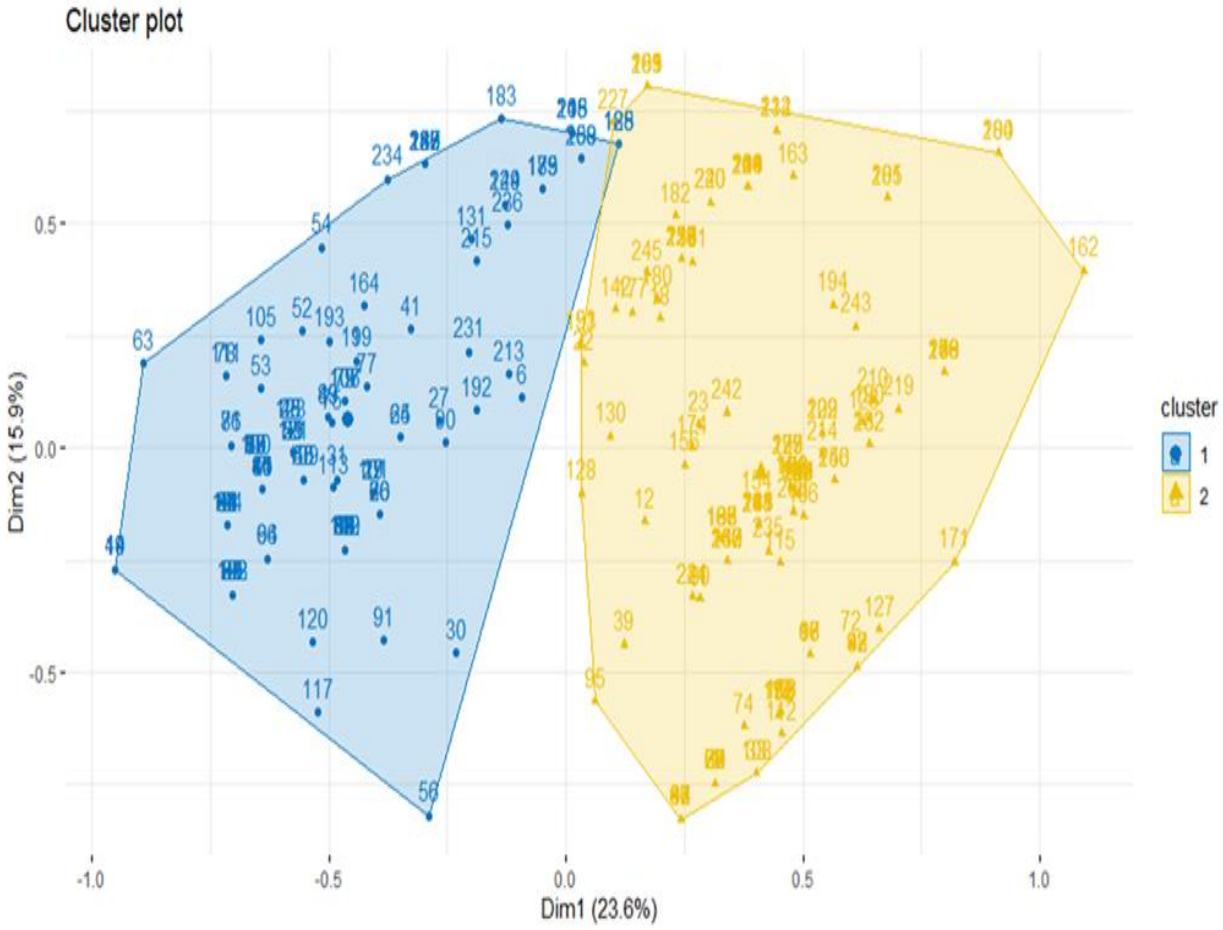


Figure 4. 17: Typology of study population regarding CL status.

The driving factors for transmission of CL in the Sodo community are follows:

- Residential kebele (Kola nurena and Beki) from demographic characteristics.
- Presence of screen o window or door (from the household factors);
- Presence of sensel tree and cave or gorge near the house (from environmental factors);
- Use of personal protective measures.
- Irrigation of crops at night, travel history; and

- Covering face and hand while sleeping, spending time near gorge/cave and sleeping at the home yard (from host-related factors) and
- Presence of hyrax and dumping animal dung near home (form animal-related factors).

4.2.3. Overview of the risk assessment survey finding

Frequencies and percentages of all possible factors in CL transmission among the Sodo community were explained in detail. These factors were subject to bivariate and multivariate logistic regression to identify their significance in disease transmission. As a result, kebele, screen on window or door, presence of sensel tree and cave/gorge near home, different host-related factors (such as travel history, sleeping at the home yard, irrigating crops at night, spending time near cave or gorge, use of personal protective measures and sleeping hand/face covered), presence of hyrax near home and in the village and dumping animal dung near home were factors significantly related with CL in logistic regression. Most of the factors affecting disease occurrence in this community are the host/behavioural factors (use of personal protective measures, irrigation at night, sleep hand/face covered, putting a screen on window and door, sleeping at the home yard, and spending time near gorge/cave) that can be altered through effective awareness creation campaigns. The multiple correspondence analysis followed by hierarchical clustering help understanding the overall relationship between selected factors and classify individuals into two distinguished groups.

4.3. PHASE THREE-RESULTS OF PREVALENCE AND SPECIES IDENTIFICATION

The third phase of the research focuses on prevalence study and parasite identification. Households were selected using a systematic method and the household head was asked for the presence of family members being actively infected with CL or about past infection. Then if a member was found to be actively infected with CL skin, slit samples were taken for microscopic examination with Giemsa staining and culture. The results of this study are described next.

In this section, clinical manifestations, the prevalence in the community, and parasites identified from the collected samples in the area will be discussing. Accordingly, the prevalence of CL in the area, demographic factors associated with prevalence, type of CL lesions and body parts affected, smear and culture test results, and findings from the PCR method will be presented.

4.3.1. Descriptive analysis of study population and related factors

The population living in the purposively selected six kebeles of Sodo District were studied. A total of 379 households and 1,356 individuals within these households who agreed to participate in the study, were included, and clinical assessment was done for individuals with a suspected case of leishmaniasis.

Case definition

Suspected case: Any person that has a single or multiple lesion that starts as a small red papule and enlarges gradually up to 2cm in diameter and forms nodules or ulcers.

Confirmed case: is a suspected case confirmed by laboratory diagnosis such as microscopy or culture (Amisalu 2014:29).

Active CL lesion: active lesions are lesions showing crater form ulcer with a raised border, plaque, smooth nodule, satellite lesion, and localized/subcutaneous nodule (Alkulaibi, Suleiman, Khalil & Al-Garadi 2019:3).

Past infection(scars): are characterized by no history of trauma, more than two weeks lesion duration, round or oval shape, smooth surface, depressed scar, and pigment change, either hypo- or hyperpigmentation (Alkulaibi et al. 2019:2).

4.3.1.1. Prevalence of cutaneous leishmaniasis and disease characteristics

Out of the 379 houses visited, 110 houses (29.0%) were harbouring at least one person positive for CL (with past or active infection). This result showed that one-third of the houses harbor an infected person indicating the endemic nature of the disease in the area. In the 379 houses, 1356 individuals were investigated and out of these,123 (9.07%) were identified with suspected cases of CL.

Disfiguring and life-long scars are formed on the face or other exposed areas because of infection from CL (Erber, Arana, Ben, Bennis, Boukthir, Castro, et al. 2020). Therefore, the presence of a scar is an indicator of infection in the past. Whereas, if the lesion is active then it means the person is infected recently. Of these, 49 (3.61%) were with active CL infection during the visit, and 74 (5.45%) were infected in the past (With scar) (Figure 4.18).

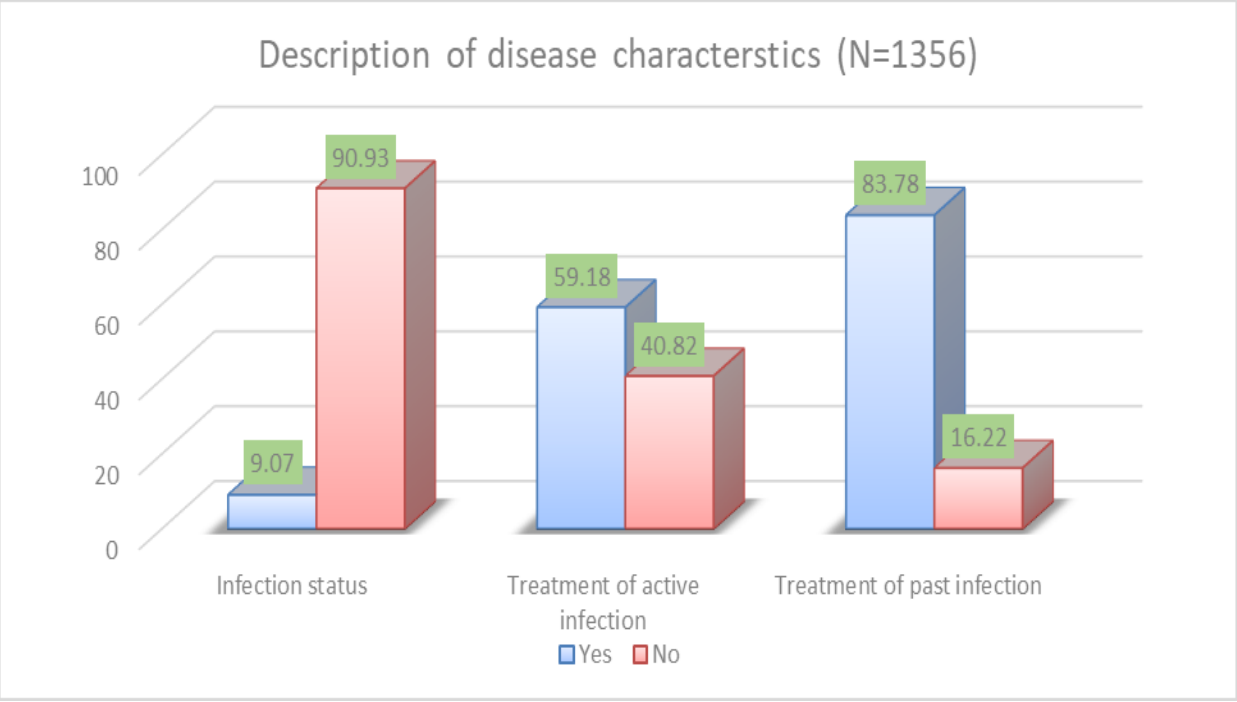


Figure 4.18: Description of disease characteristics (n=1356).

More than half (59.18%, 29/49) of people with the active infection get it treated. The treatment method used by 72.41% of treated patients with active cases was the application of herbal medicines. Of the 62 (83.78%) of the participants with past infection who seek treatment, the majority (82.08%) visited herbal healers and only seven patients (11.29%) go to modern health facilities seeking treatment. When asked who in the family was infected in the past, 47 (63.5%) indicated as self, and 27 (36.48%) said that it was another member of the family.

4.3.1.2. Socio-demographic features of persons with cutaneous leishmaniasis

Table 4.23 describes the demographic characteristics of the study participants with suspected CL. Looking into the kebele in which positive cases belong, 34 (34.69%) were from Kola Nurena kebele. Females were with a slightly higher percentage of suspected CL (9.49%) than males (8.6%) in this study.

Illiterate individuals (10.35%) and who are only able to read and write (10.81%) are the ones holding a bigger proportion of the suspected cases. In the current study, the proportion of CL positivity was slightly higher (9.85%) in respondents whose occupation involves farming than those who have a non-farm related job (8.39%).

Considering the age, out of the 123 with CL 10 were between 5-10 years, and all (100.0%) of them were positive (eight with active CL and two with scar). Flowing 5-10 years, 25-32, and 35-44 years were found with CL.

Table 4.23: Demographic characteristics of individuals with suspected CL.

Variable	Total sample(n=1356)	Number of CL positive(n=123)	% CL positive
Kebele			
Adazer	509	34	6.68
Beki	71	11	15.49
Genete Mariam	326	16	4.90
Kela Zuria	171	9	5.26
Kola Nurena	98	34	34.69
Michael Semero	181	19	10.50
Gender			
Male	661	57	8.6
Female	695	66	9.49
Education			
Illiterate	454	47	10.35
Able to read and write	185	20	10.81
Primary education	422	38	9.00
Informal education	21	1	4.76
Secondary and above	269	17	6.32
Occupation involving farm			
Yes	619	61	9.85
No	739	62	8.39
Age			
5-10	10	10	100.00
10-17	306	21	6.86
18-24	183	13	7.10
25-32	259	28	10.81
35-44	295	29	9.83
45-54	172	9	5.23
>54	121	13	10.74

4.3.1.3. Type of cutaneous leishmaniasis and body parts affected by active lesion

One hundred and twenty-three (9.07%) of the participants were infected with the parasite at present time or in the past. The total prevalence of active CL lesion in the study population was 3.61% (49/1,356). More than half (59.18%) of those with active CL used herbal medicines. Details of clinical presentations and the type of CL observed in participants with active lesions are presented in Table 4.24.

Table 4.24: Clinical futures of confirmed CL cases (n=49).

Confirmed cutaneous leishmaniasis	Frequency (%)
CL type	
LCL	43 (87.79%)
MCL	6 (12.24%)
Lesion number *	
1	41(83.67%)
2	5(10.20%)
3 or more	3 (6.1%)
Duration of lesion **	
Less than 7 months	16 (32.65%)
7 months to 1 year	20 (40.8%)
Above one year	13 (26.5%)
Location of lesion *	
Cheek	14 (28.57%)
Ear	4 (8.1%)
Lips	5 (10.20%)
Forehead	2 (4.08%)
Hand	3(6.12%)
Chin	3 (6.12%)
Nose	7(14.28%)
Neck	2 (4.08%)
Multiple sites	
cheek, chin, nose	2 (4.08%)
cheek & nose	3 (6.12%)
cheek, ear	1(2.04%)
Lip, Nose	1(2.04%)
Cheek, Hand	2(4.08%)

*= obtained by clinical assessment, **= obtained by interviewing the patient

Among the active CL, cases confirmed clinically, the majority 87.79% (43/49) were presented with LCL and the rest 6 (12.24%) were found to have MCL. LCL could heal over time or may damage surrounding tissues but the significant issue with MCL is that the parasites spread to nose, throat, and mouth causing partial or complete damage of the mucous membranes (SastaSundar 2020).

Considering the number of lesions on an infected person, most of the patients, 41 (83.67%) had single lesions and only 3 (6.1%) developed three or more lesions.

Leishmaniasis has variable incubation period ranging from two weeks to several years. However, most individuals develop symptoms after about two to six months

(Davis 2019). The duration of the lesion was assessed in this study by questioning the patients, and the majority of them, 20 (40.8%), had a lesion that began between seven months and one year ago. The lesion began in less than seven months for sixteen (32.65%).

In terms of the lesion's site, which was determined by physical examination, more than half of the CL cases (28.57%) acquired a lesion on their cheeks. Lesions on numerous sites, in addition to the cheek, were typical 9 (18.36 %). Face parts (nose, ear, lips, cheeks, forehead, chin) were affected by 89.79% (44/49) of the cases.

CL can take various lesion natures based on the parasite species, location of the lesion and many other factors. An open lesion that is eroding the skin or mucous membrane is called an ulcer. Nodules are a small collection of tissue that can be felt during palpation at any level of the skin from epidermis to subcutis or in another tissue of the body (Handler, Patel, Kapila, Al-Qubati, Schwartz 2015). Different lesion nature types were observed in the present study, such as ulcer 20 (40.81%) and nodules with or without crust 22 (44.89%). Most of the lesions were dry 36 (73.46%) and the remaining 13 (26.53%) were wet. Of the wet lesions, seven (14.28%) of the lesions were exudative and draining and others were painful with pruritis.

4.3.1.4. Smear test result

Smear test using Giemsa bloodstain is a dye utilised to identify parasites in blood or tissue samples. Giemsa stain identifies parasites pink or red a color which is different from human blood cells. Furthermore, Giemsa stain binds with DNA within human cells and stain these with purple color. Skin scrapings from a total of 49 participants who had active lesions and consented to take samples were smear tested and pink amastigotes were visualised from 34.69% (17/49) of them.

Table 4.25: Characteristics of cases positive for a smear test (n=17).

Characters	Frequency (%)
Age	
5-10	8(47.05)
11-20	4(23.52)
21-30	3(17.64)
>31	2(11.76)
Sex	
Male	7(41.17)
Female	10(58.82)
Form of CL	
LCL	14(82.35)
MCL	3(17.64)

Six (35.29%) of the 17 smear-positive cases received herbal treatment. Logistic regression was used to identify a significant difference in test positivity (both smear and culture) between individuals who used and did not use herbal treatment. Bivariate logistic regression test of receiving herbal treatment and smear test revealed that those who received herbal medication previously are less likely to have positive results in smear test [COR= 0.21; 95% CI, 0.057-0.726] and culturing [COR= 0.27; 95% CI, 0.050-1.182] (Table 4.26).

Table 4.26: Logistic regression of herbal treatment and test results.

Test	Univariate	
	p-value	**COR (95%CI)**
Smear		
Negative		
Positive	0.0161 *	0.21(0.057-0.726)
Culture		
Negative		
Positive	0.0929.	0.27(0.050-1.182)

Of the 17 smear-positive cases, 14 were showing LCL from and the rest MCL. Pictures of clinical lesions of LCL and MCL from the current study population are seen in Figure 4.19 and Figure 4.20, respectively. These pictures are taken from individuals who consented their picture to be taken. From the 14 smear-positive cases taking the form of LCL, only two (14.28%) were with a history of travel.

Figure 4.19: Some of the lesions taking LCL form.

A. Male 6 years old lesion on the right eyebrow with a one-year duration. B. Male 7 years old lesion on the left chin with a six-month duration. C. Female 6 years old with lesion duration of one year. D & E. Both from single person female 11 years old lesion on the right cheek and right arm with a duration of one year. F. Female 9 years old with a lesion of one-year duration. G. Three years old boy with lesion under the chin of one-year duration. H. Three years old boy with lesion under the chin starting from five months. I. 26 years old male with a lesion on nose starting five months.

All three MCL lesions occur on the lower lip as shown in Figure 4.20. None of these MCL cases has a history of travel and one patient (with a picture at the right side) confirmed treatment of the lesion with herbal medicines.

Figure 4.20. Lesions with MCL form.

Right side: 10 years old girl with lesion duration of 6 months. Left side: 28 years old female with a lesion duration of 15 days.

4.3.1.5. Culture results

From 49 cultured samples, 18.37% (9/49) were culture positive and 81.63% (40/49) were culture-negative in Novy-MacNeal-Nicolle (NNN) medium.

Out of the nine culture-positive samples in the current study, two (22.2%) were contaminated so deemed unfit for polymerase chain reaction (PCR) analysis. As well-known with all cultures, Novy-MacNeal-Nicolle (NNN) or other Leishmania medium are also exposed to the contamination problem (Nuako 2016:41).

Looking into the clinical forms of nine culture-positive samples two were from patients who have MCL and the rest seven were from patients with LCL. Of the nine samples positive for culturing on NNN medium of skin scrapings, three (33.3%) have received herbal medication this shows that herbal remedies may remove parasites from the site but do not completely heal the person.

4.3.1.6. PCR amplification with the genus-specific kDNA-primers 13A/13B

In this investigation, the genus-specific primer pair 13A/13B was used as a screening strategy. The PCR result was judged positive for the Leishmania genus when a band of the expected size (120 bp for 13A/13B) was obtained. From promastigotes, the preserved 120 bp size miniexon KDNA was amplified (culture). All nine culture samples evaluated with PCR for the presence of Leishmania amastigote DNA were found to be positive (Figure 4.21).

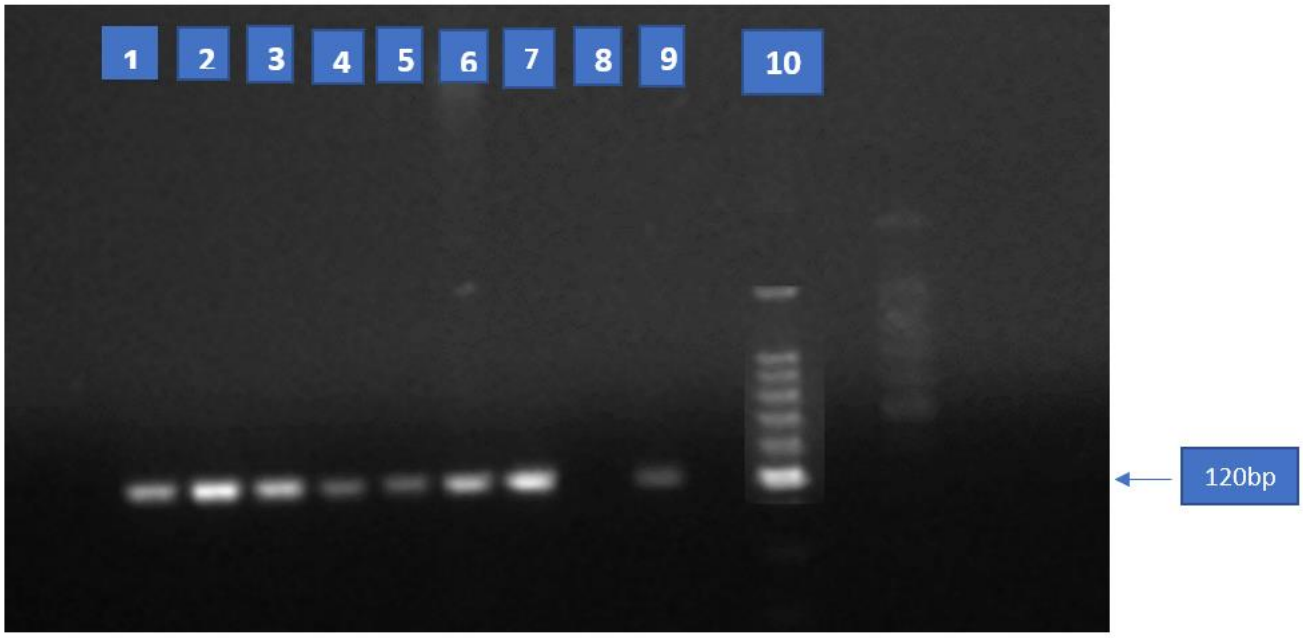


Figure 4.21: Images of PCR product amplified with primer pair 13A/13B. After staining with ethidium bromide from **culture promastigotes**: 1-7 Clinical samples, 8= Negative control (TE),9= Positive control (*L. aethiopica*), 10= 1kb ladder.

4.3.1.7. PCR-Restriction Fragment Length Polymorphism (RFLP) analysis

The restriction fragments were examined using 2.5% agarose electrophoresis and ethidium bromide staining, and UV light was used to visualise them. When the ITS-1 PCR result was digested with the enzyme Hha I, the patient samples and the *L. aethiopica* reference strain produced superimposed products of 164 and 162 bp, showing that all of the CL cases were caused by *L. aethiopica* (Figure 4.22).

The isolates discovered from the current study sites are L. aethiopica, with an RFLP pattern comparable to that of the *L. aethiopica* reference strain. This finding supports the hypothesis that *L. aethiopica* is the primary cause of CL in Sodo.

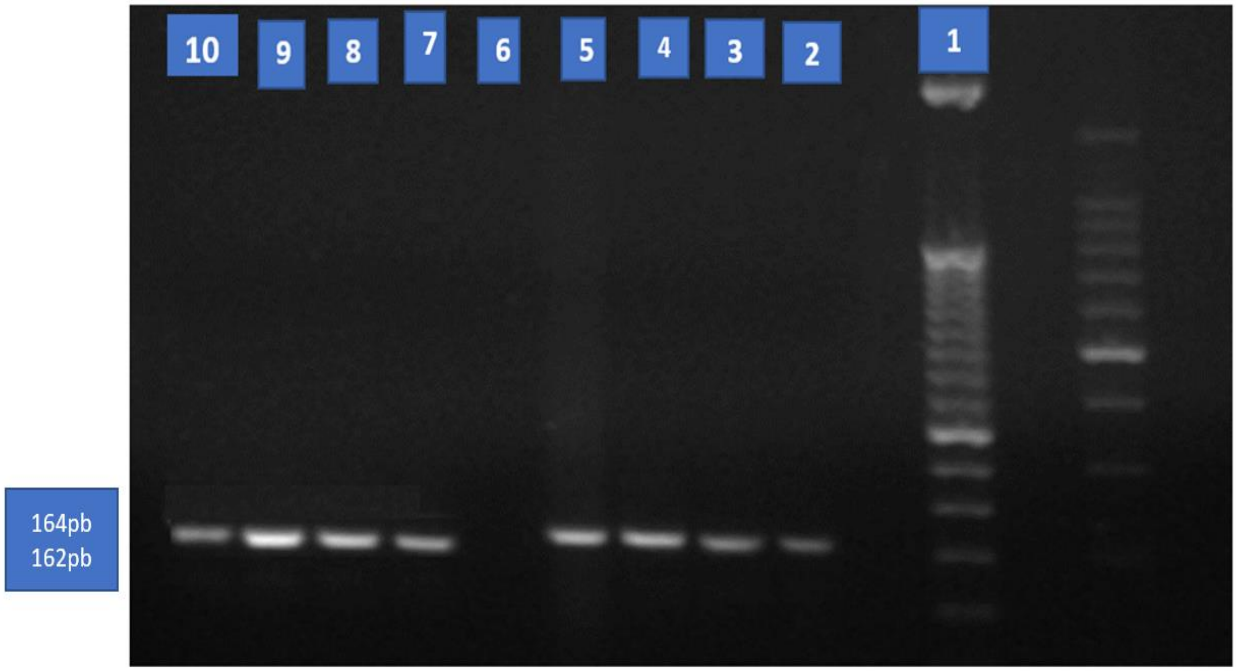


Figure 4.22: PCR-ITS1-RFLP of the amplicon with *Hha I* from culture promastigote.

6= Distilled H2O is the negative control; 9= *L. aethiopica* issued as a positive control, 1=A 1 kb ladder is the molecular size marker. 2,3,4,5,7,8 & 10 are clinical samples.

4.3.2. Overview of the research finding

In this section, efforts made to estimate CL prevalence and identify the parasite are described in detail. Accordingly, the clinical assessment was made on the 123 patients with active infection and with a scar. Here demographic factors associated with the disease are discussed. Samples collected from 49 patients with active cases were subjected to microscopic examination through the preparation of smear by Giemsa staining and NNN medium. Finally, PCR amplification with the genus-specific kDNA-primers 13A/13B for genus identification and PCR-Restriction

Fragment Length Polymorphism (RFLP) analysis for species identification were applied and results presented.

4.5. CONCLUSION

In this chapter, the result of the community's knowledge attitude, and practice were presented. Socio-demographic factors influencing knowledge attitude and practice have been analysed through the application of chi-square test, bivariate and multivariate logistic regression. Factors playing major roles in the transmission of CL in the community were successfully identified, meeting the objective set in the methodology section and producing information that will help decision making for designing effective and less costly strategies to prevent the disease and enhance community health. The overall prevalence of CL in the area was 9.07% (123/1,356) for both scar and active infection. Of these, 49 (3.61%) were with active CL infection during the visit, and 74 (5.45%) were infected in the past (With scar). There is a difference in the proportion of active CL between different residential kebeles, gender, age and educational status. The most generic form of CL among cases was LCL and lesions are mainly located on face parts. This result offers proof for *L. aethiopica* to be the primary causative agent of CL in Sodo which is the first report in the area to the researcher’s knowledge.

In the following chapter, discussions of the results compared to other research findings will be presented.

CHAPTER 5

DISCUSSIONS OF RESEARCH FINDINGS

5.1. INTRODUCTION

The quantitative data analysis and study outcomes were introduced in the previous chapter. In chapter 4, the results of the KAP survey, risk factor analysis, and prevalence and species identification were all detailed. This chapter reviews the thesis' results in accordance to the design of the study. It also discuses the findings in comparison to previous surveys, existing policies and strategies.

5.2. DISCUSSION OF PHASE ONE – THE KNOWLEDGE ATTITUDE AND PRACTICE SURVEY

Here findings in knowledge, attitude and practice survey are discussed in comparison to similar studies in Ethiopia, Africa, and other parts of the world. Factors affecting knowledge attitude and practice are also discussed in comparison to other research findings below.

5.2.1. Socio-demographic characteristic of the respondents

A total of 423 household heads were involved in this study. The proportion of male respondents was slightly higher than females. About 60.0% of the respondents are above 24.5 years old. This may be because household heads are interviewed in most cases and any elder person in the absence of household heads in a few cases. Unlike the many districts in southern regions of the country where protestant religious worshipers are mainly concentrated, Orthodox was the dominant religion in Sodo District. However, this agrees with the national census where Ethiopian Orthodox is a religion to 43.5% of Ethiopians (Central Statistical Agency 2014).

Participants' major occupation was farming [216 (51.1%)] and 188 (44.4%) of the participants claim to be housewives. Sodo is mostly a rural area. As a result, there is a minor difference in occupation type among residents as most depend on on-farm activities. Those who said their job is being a housewife have a spouse who is a farmer, or they make their livelihood on poultry, vegetables, or other backyard farm activities.

Within the study participants, the average family size was 5.76. In Ethiopia, the average family size is anticipated to be 4.7, which is higher than the national average (CSA 2014). The average duration of stay in the district was 32.79 years. Accordingly, only a few proportions of the study participants 81 (19.1%) came from another place while the rest 342 (80.9%) are originally from the same district. Considering the education level of the respondents, the majority are illiterates or only able to read and write only.

5.2.2. Knowledge of the community about cutaneous leishmaniasis

In the current study, 216 (51.1%) of the participants identified the case of CL after shown a picture illustrating cases of CL reported from Ethiopia and 350 (82.7%) of the participants have heard about CL. A lower proportion (39.8%) of inhabitants of the Kani forest tribal settlements can recognise CL when shown a picture of CL manifestation (Nandha et al. 2014:1). Similar studies also indicate that 10% (In West Alexandria) (Randa, Naguiba & Osama 2015), 29.0% (in Paraguay) (Ruoti, Oddone, Lampert, Orué, Miles, Alexander, Rehman, Njord, Shu, Brice, Sinclair & Krentel 2013:3) and 47.9% (in Southwestern Iran) (Vahabi, Rassi, Oshaghi, Vahabi, Rafizadeh & Sayyad 2013:865) were aware of the disease and can recognise CL lesions. Higher percentage reports of awareness CL lesions 75.0%, 82.0%, and 85.9% were seen from Nigeria among physicians in health care facilities (Awosan et al. 2013:258), from communities in Ghana (Doe, Egyir-Yawson, Kwakye-Nuako 2019:35) and communities in Colombia respectively (Pardo et al. 2006). Even majority (91.7%) of the indigenous community in French Guiana and Brazil were able to recognise a photograph with classic case of CL from French Guiana (Odonne, Berger, Stien, Grenand, & Bourdy 2011:1231) and most of the respondents (81.0%) in Syrian Arab republic know at least one local name of the disease (Abazid et al. 2012:10). The difference between these records and the current study may be because these studies were conducted among individuals who are health professionals (Nigeria), among people during CL occurrence were highest (WHO 2016) in the country (Colombia) and among the most ancient tribal of

French Guiana who has been facing and managing CL by themselves without the intervention of modern medicine.

In the present study, majority of study population 350 (82.7%) have heard about CL. This was in line with a study from Southern Iran, where 83.0% (Sarkari, Qasem & Shafaf 2014:567) of participants had heard about CL before. Similarly, in the current study, 82.7% (n=350) of the respondents had heard about CL. This figure was greater than the findings of an Ethiopian research, which found that 265 people (67.6%) had heard about CL (Kebede et al. 2016:565). In comparison to a Paraguayan study (Giménez-Ayala, Brtez, Rojas-de- Arias & Ruoti 2018), which found that 71.0% of participants had heard of CL, the current study's finding is greater. However, it is lower than those from other research, which show that 97.3% and 92.2% of participants in Sri Lanka and Iran, respectively, have heard about CL before. (Weerakoon, Ranawaka, Bandara, Herath & Warnasekara 2016:8, Hejazi et al. 2010: 36). This disparity could be due to the fact that the other studies were conducted among health professionals (in Sri Lanka) and the difference in educational status of mothers of CL-affected children (where only 6.2% are illiterate) and the current study (where only 6.2% are illiterate) (where 51.1% are illiterate).

Regarding the source of disease information for participants in the current study, the majority of 350 (82.7%) have heard about CL primarily from families, friends, and neighbours and (67.5%, n=286) from colleagues and (9.0%, n=38) being exposed to the disease. Sources of information on CL for 82.7% of the participants were families, friends, and neighbours. This finding is similar to a study from Paraguayan communities which reported (57%, 143/250) of the population gain information about CL from their family, friend, or neighbour (Ruoti et al. 2013:3). Similarly, in Pakistan, friends, neighbours, or teachers were the source of information for majority of the population (Akram, et al. 2015:6). Also, most of the participants (95.5%) in Ghana that had heard of the disease, had first-hand knowledge from the community in which they lived (Doe et al 2019:35). It has been indicated that the mass media have various functions in society and can help develop the community (Khosravani, Ghavam & Yazdan 2016:63). In the current study, Professionals like

Health workers, religious leaders, schoolteachers, and teaching tools like media, Brochures, and newsletters which are believed to be vital gear for community health awareness creation program collectively share a small percentage 38 (9.0%) of the knowledge attained about the disease. Similar reports were seen in Kharameh about the low performance of awareness creation through radio and television on preventive behaviours of CL (Khosravani, Ghavam & Yazdan 2016:63). The use of teaching and electronic media for designing leishmaniasis control can be effective (Akram, et al. 2015:6). Unlikely in Iran 53.4% of participants had mentioned that their first premiership for gaining the information about CL was "Health-workers" (Maryam et al. 2016). Television (38.0%, 96/250), radio (30.0%, 76/250), community health worker (12.0%, 29/250), school (6.0%, 15/250), and brochure or poster (1.0%, 3/250) were the top sources of illness knowledge in Paraguay (Ruoti et al. 2013:3). In Sri Lanka, nearly half of the respondents (48.2%) had participated in awareness programs, and 45.8% had gained further knowledge by reading pamphlets, 25.6% by watching television, and 25.7% by reading newspapers (Weerakoon et al. 2016:8).

In this study, most 301 (71.2%) of the respondents knew someone infected with CL in their area. Likewise, a study in Nigeria revealed that 79.3% of the study participants in Nigeria reported ever seeing a person with a CL case and this, as the author explains, resulted in better recognition of CL lesions (Awosan et al., 2013:259). In contrast to the above reports, the percentage of respondents seeing the case of CL before was lower in Pakistan (24.4%) (Akram et al. 2015:4). Much lower (8.3%) population of the local inhabitants in Egypt know a previously diseased person (Randa et al. 2015:2171). This difference may be because larger populations were considered in the current study compared to other studies in Pakistan (250 participants) and Egypt (60 participants). Since the report from Nigeria was obtained from 164 doctors, it is known that they would probably see patients with CL more frequently. One hundred and fourteen (68.7%) mothers of children affected by CL had a positive family history of leishmaniasis (Hejazi et al. 2010:36). Similarly, in Southern Iran 63.0% of the respondents have seen CL infected person or have an infected family member (Sarkari et al. 2016:568).

According to the respondents in the current study, the individuals know of neighbours, family members, cousins, and self who are infected. This may suggest the local transmission of the disease. In Ghana, 44.1% of the participants experienced the disease, 59.5% had family members who had the disease in the past and 71.2%, knew unrelated people who had experienced the disease (Doe et al. 2019:37). Among Sir Lanka health workers, 12.5% were victims of the disease and in 3.75% of them, the disease occurs in their family and in 8.5% cases their neighbours were infected (Weerakoon et al., 2016:8) and 68 (51.5%) of the Wayãpi Indians had suffered from the disease previously (Odonne et al. 2011).

In the current study, 206 (53.4%) of the respondents do not know the cause of CL. Another 252 (59.4%) do not know ways of disease transmission. This indicates that most of the study participants in Sodo area have little or no knowledge of CL cause but a higher proportion of participants (80.0%) in Ecuador knew how CL is transmitted (Sorocco et al. 2017:165). Similarly, in Southern Iran, more than half of the respondents informed that microbe cause CL, and even a few 1.2% name Leishmania parasite as the causal agent of CL (Sarkari et al 2014:567). Sixty-six of the respondents in the Syrian Arab Republic know the possible cause of the disease mentioning sandfly, mosquitoes, water, and finally microbes (Abazid et al 2012:10). In Sri Lanka 97.75% of the study population nhave good understanding of CL transmission (Weerakoon et al. 2016:8) and as little as, 19.8% of the participants of the endemic communities in Ghana knew that a fly (insect vector) transmits the agent causing the disease, but they are unable specify sandflies. On the other hand, 80.2% of those polled had no understanding how CL was transmitted (Doe et al. 2019:35).

The current study revealed that plaque and pus-containing wounds were mentioned as the main clinical symptom of CL by 268 (63.4%) and 206 (48.6%) of the respondents, respectively. On the contrary, 92 (21.7 %) confess that they are not aware of any CL symptoms. Knowledge of the symptoms and indicators of CL can aid individuals and their families in avoiding the disease (Rassi, Yavar & Saghafipour, Abedin & Abai, et al., 2011). Skin infection, fever, enlargement of the

liver and spleen, and anemia are the most common leishmaniasis symptoms in Pakistan, according to 33.6%, but the majority (56.0%) are ignorant (Akram et al. 2015:4). In endemic locations, there is a lack of knowledge about the disease and its vector (Vahabi et al. 2013:866). Thirty percent of the Paraguayan population is unaware of any sickness symptoms (Ruoti et al. 2013:3). According to a study conducted in Ethiopia, 59.6% of participants recognised swollen legs as a prominent indication, whereas 11.0% were unable to name any sign (Kebede et al 2016:565). The majority (95.8%) of students at Airbase Isfahan knowledgeable about signs and symptoms of CL (Saberi, Zamani, Motamedi, et al. 2012:308). With regards to the symptoms associated with leishmaniasis, 48.7% of the respondents in Ghana reported that it was associated with itching and painful swelling sore, 34.2% reported only deep painful sore and 17.1% reported they had no idea (Doe et al. 2019:35). Plaques and papules were identified as common skin manifestations by more than 70.0% of Sri Lankan health workers, but skin rash was identified by just 16.5% (Weerakoon et al. 2016:8). In Guatemala, 96.7% of the 425 heads of families asked could appropriately describe a typical CL lesion. (Arana et al. 2000).

Nose (316 (74.5%)), face (305 (71.9%)), forehead (116 (27.4%)) and ear (102 (24.1%)) are parts of the body mentioned by the respondents to be mostly infected by the lesion (called “kuselet” in Amharic) from CL infection. Similarly, the majority (59.7%) of the participants in South Ethiopia answered that lesion appearing on face was major sign of CL infection (Kebede et al. 2016:565). This group also stated that the leg (51.5%) was the most prevalent location for scars, followed by the thigh (11.8%) and the arm (8.8%) (Kebede et al. 2016:565). Legs (16.3%), forearms (13.8%), thorax (13.3%), and face (10.4%) were the most commonly reported sites of leishmaniasis ulcers at Cayenne Hospital (Klisnick, 2006: in Odonne et al 2011:1233). It is obvious that these parts of the body are usually left uncovered thus are easily assessable for vectors A major fraction (95.3%) of health workers in Sri Lanka knew skin is the frequently affected body part (Weerakoon et al. 2016:8).

In this study concerning knowledge of the participants about the vector sandfly, nearly half 210 (49.6%) reported to know the biting and blood-sucking behaviour

after being shown a picture of phlebotomine sandfly collected from nearby districts. In the Tigray Region of Ethiopia, 81 (47.1%) of the respondents were able to recognise sandfly, with 67 (82.72%) of them knowing at least one sandfly resting/breeding site (Berhe, Bsrat, Taddele, et al. 2018:4). These indicate a low level of awareness regarding the sandfly vector.

Even though sandflies are important leishmaniasis vectors, few people were aware of their role in infection. Lack of information about CL and its vectors is an issue for implementing prevention measures and seeking early treatment (Akram et al 2015:4). Respondents from Paraguay (29.0%) (Ruoti et al. 2013:3), Pakistan (27.6%) (Akram et al. 2015:4), and Southwestern Iran (39.5%) (Vahabi et al. 2013:865) all believed that sandflies were responsible for the disease's spread. Many Nigerian physicians at tertiary health care facilities were aware that leishmaniasis is transmitted through the biting of infected phlebotomine sand flies (Awosan et al. 2013:260). This demonstrates the possibility of boosting community awareness through the involvement of health care workers, as when health care workers are self-aware, a good environment for enhanced caring is produced, opening the door to personal achievement and proper management. In addition to the poor awareness about sandfly vector, there seems to be also confusion between sandfly and mosquitoes as the vector of leishmaniasis. About 77.2% of these are from Ethiopia (Kebede et al. 2016:565), 37.5% from India (Nandha et al. 2014:1053), 25.0% from Nigeria (Awosan et al. 2013:260), 8.0% of the communities from Paraguay (Ruoti et al. 2013:3) and 63.5% of the South Iran communities (Sarkari et al. 2014:566) said that mosquitoes are responsible for the transmission of the disease. Even though control strategies for sandfly and mosquitoes are similar, there is a difference in breeding sites and biting habits of these vectors. Therefore, strategies for reduction of sandfly – human contact and sandfly population surrounding households must be designed differently from control measures for mosquitoes. For the community to actively engage in such preventive strategies, awareness creation on the identification of sandfly, their breeding, biting habits, and resting sites is of vital importance.

Previous investigations in Ethiopia revealed that nearly 20% of study participants had no understanding how the disease was transmitted, and of those who did, 192 (49.0%) said sandfly bites were the main mode of transmission, while 63 (16.1%) said flies were the main mode of transmission (Kebede et al. 2016:565). No knowledge of the Leishmania vector was also reported elsewhere as 37.3% of participants from Southwestern Iran (Vahabi et al. 2013:865) and 57.6% from Pakistan (Akram et al. 2015:4) did not have any knowledge of the vectors of the disease. When shown live sandflies gathered from their homes, some respondents had no idea about the biting and blood-sucking activity of sandflies, and 65.0% mistook sandflies for fruit flies (Drosophila melanogaster) (Nandha et al. 2014:1054). Because of this lack of awareness, preventive methods may fail, making education about this vector essential (Hejazi et al. 2010:39).

As little as 19.8% of the communities in endemic areas of Ghana knew that a fly (insect vector) transmits the agent causing the disease. However, they could not confirm this to be the sandfly while 80.2% of the participants had no idea about the mode of transmission of the disease (Doe et al. 2019:35). A shortage of knowledge about the sandfly vector was also found in Colombia (Patiño-Londoño, Salazar, Acero, Bernal 2017:1). The most cited etiology of leishmaniasis in French Guiana was an insect bite (such as ants, horseflies, mosquitoes, and wasps) and a wound (Odonne et al. 2011:1231).

The vector's preferred breeding area and biting time were unknown to 123 (29.0%) and 139 (32.8%) of the participants in the current study, respectively. The vector bits were mostly visible at nightfall, according to 178 (42.0%). Sandflies are most active throughout the evening, nocturnal, and twilight hours (from dusk to dawn), and are less active during the hottest hours of the day. If they are disturbed, though, they will bite (MedicineNet 2007). The most commonly stated breeding areas of the vector include dirty places (218 (51.5%), water ponds (194 (45.9%), and waste collection sites (111 (26.2%). Cervices in the house (18.3%), thatched roof (17.4%), and cattle shades (20%) all received a little percentage of the vote (4.7%). Sand flies' immature stages are more concentrated in microhabitats with specific

characteristics, such as the presence of organic materials, wetness, and low light levels (Sangiorgi, Miranda, Oliveira, et al. 2012:2).

Knowledge of the breeding and resting places of the vector is important disease preventive method. If the communities in the endemic areas are made aware of these places, they could reduce vector-human contact by denying access to breeding and resting places within the household. In Ethiopia, 77.3% of the participants indicated that sandflies breed in the vegetation (Kebede et al. 2016:566). Different breeding grounds for sandflies were cited in Pakistan, with 18.4% believing that filthy conditions are good breeding places, while 10.8%, 6.4%, and 4.8% saying wet places, freshwater, and hospitals waste disposal sites, respectively. However, 59.6% of respondents said they didn't know where sand flies breed (Akram et al. 2015:3). According to another survey, a large majority of Iranian respondents (46.5%) had no clue or were incorrect (39.5%) regarding the vector's breeding site, with most individuals claiming unclean regions, water ponds, and trash gathering sites as the greatest likely breeding sites for the vector (Sarkari et al. 2014:568).

In the same Pakistani survey, 14.0% of respondents believed that sandflies bite at dusk and down, followed by at any time of day (13.2%), during the daytime (10.0%), and at midnight (8.0%), while the majority (54.8%) were uninformed (Akram et al. 2015:3). In India, 42.5% of people were unaware of the disease's transmission. Furthermore, the majority of the patients (67.5%) lacked knowledge of sandfly breeding sites and biting times (75.0%). (Grapati, Pal, Siddiqui, etal.2018:5). Due to a lack of knowledge regarding sandfly biting times in endemic areas, personal protection measures such as mosquito nets, insecticides, and mosquito repellent may be used less frequently during those times. A case control study in Nepal demonstrated the effectiveness of employing bed nets for prevention in VL (Bern, Joshi, Jha, et al. 2000).

5.2.3. Discussion of factors affecting knowledge of the study participants

Binary logistic regression (for both bivariate and multivariate analysis) was used for evaluation purposes and see effect of variables on on participants' knowledge, attitude, and practice about CL. In bivariate logistic regression, females are knowledgeable than males [COR=2.9; 95% CI, 1.919-4.371] in the current study. Accordingly, females are three times likely to have satisfactory knowledge than males. Similarly, the total knowledge about leishmaniasis and sandfly was satisfactory in 9 (60.0%) of the females and 2 (40.0%) of the males of the West Alexandria primary health physicians which is this result is statistically insignificant ($p < 0.617$) (Randa et al. 2015:2168). Another study also showed that there was a significant statistical variation between awareness about the disease and gender (Vahabi et al. 2013:865). Girls were also more knowledgeable CL ($p < 0.001$) among students at Shahid Babaie Airbase (Saberi et al. 2012:309). According to a study conducted to analyse gender discrepancies in health information practices, men are often cautious and lack the inspiration to engage with health-related information (Stefan 2015:1). In contrast to these findings, another Ethiopian study indicated that 153 (68.0%) of 225 males who heard about CL were knowledgeable, while 112 (67.1%) of 167 female participants were (Kebede et al. 2016:565). Housewives, on the other hand, are more knowledgeable than farmers [OR=1.627; 95% CI, 1.097-2.412]. This may be because housewives are females.

When compared to illiterate respondents, people who can read and write are less likely to be knowledgeable [COR=0.49; 95% CI, 0.310-0.771]. This means there is no substantial difference between illiterates and who can read and write for acquiring knowledge of CL. Respondents with a primary education, on the other hand, are more likely than illiterate respondents to have a better understanding of CL [COR=2.5; 95% CI, 1.238-5.105]. There was a statistically significant difference between disease awareness and education level ($df=4$, $p=0.007$) (Vahabi et al. 2013:865). Similarly, there was a strong association between their degree of education and their knowledge of CL ($p < 0.001$) at Shahid Babaie Airbase (Saberi et al. 2012:307). In Southern Iran, a statistically significant link between literacy and awareness was likely discovered (Sarkari et al. 2014:568). Better education can

lead to a better chance of acquiring correct information, therefore having better knowledge. The mean scores of knowledge, attitude, enabling factors, behavioural intentions, and behavior showed a significant increase two months after educational intervention ($p < 0.05$) in a study aimed to evaluate beliefs, attitudes, subjective norms, and enabling factors (BASNEF) model in Ethiopia (Saghafipour, Nejati, Mozaffari, et al. 2017:1). However, the outcomes of this survey revealed that the majority of participants were either illiterate or had a low level of education, with 216 (51.1%) being illiterates and 119 (28.1%) only being able to read and write. Therefore, the level of education is not a good discriminatory factor in this study as nearly all (335) participants fall in the same category. However, there was no significant association between education and knowledge in mothers of children affected by CL (Hejazi et al. 2010:37). This may also indicate that knowledge-acquiring mechanism within the community is through non-modern knowledge transfer as illiterate ones are found to have satisfactory knowledge of the disease than those who can read and write.

Individuals who are not originally from Sodo District are knowledgeable compared to participants who come from other places [COR=1.84; 95% CI, 1.075-3.136]. This shows that knowledge of the population in the current study is lower than those elsewhere in Ethiopia. In a KAP study conducted by Kebede et al. (2016) in Ochollo, South Ethiopia, the overall knowledge of the community was good for 265 (67.6%) compared to 261 (61.9%) satisfactory knowledge of the population in this study.

Study participants who claim not to know a person infected with the disease are less knowledgeable than those who do [COR=0.156; 95% CI, 0.098-0.248]. Similarly, 111 (72.5%) of the 153 moms who had heard of CL had a positive family history of the disease. This study also found a link between familiarity with leishmaniasis and a positive family history of the disease (Hejazi et al 2010:36). Furthermore, in Iran, the history of CL has a tremendous impact on people's knowledge (Rakhshani et al. 2017). According to a study conducted in Saudi Arabia, a considerably higher knowledge score was associated with previous CL

history (Amin, Kaliyadan, Al-Ajyan, et al. 2012). Age was shown to have no statistically significant relationship.

From results in the chi-square test and logistic regression, it can generally be concluded that after adjusting all variables, receiving primary education, and knowing someone infected with CL are factors that truly contribute to satisfactory knowledge of CL. In Iran household size was a stronger of awerness in results of the multiple linear regression analysis (Rakhshani et al. 2017:188).

5.2.4. Attitude towards cutaneous leishmaniasis

Two hundred sixty-one (61.7%) of the participants in this study believed that CL can be treated. This attitude is important for seeking treatment and reduce the disease prevalence. However, 70 (16.5%) of the participants think that CL is not treatable and 85 (20.1%) have no idea whether the disease can be treated or not. A larger proportion (70.4%) of respondents in Pakistan thought that the disease is curable (Akram et al. 2015:4). In contrast, in Ethiopia, the majority of participants (54.8%) believe CL is not treated, and 34.9% do not know if CL is treatable (Kebede et al. 2016:567). Similarly, only 48.0% of people in Southern Iran believe CL can be treated with medicine (Sarkari et al. 2014:568). If people in a community have a positive attitude toward CL treatment, they are more likely to take steps to avoid it from happening to them or to visit a healthcare professional for regular check-ups. On the other hand, communities with a negative attitude toward treatment may choose to forego adopting preventative measures or visiting a doctor and seeking therapy.

In this study, more than half 264 (62.4%) of the study subjects think CL infection causes disfiguring and 67 (15.8%) consider CL infection to result in both disfiguring and death while 54 (12.8%) have no idea on the outcome of CL infection. More than half (57.4%) of participants included in a similar study conducted in other parts of Ethiopia had the attitude that CL had a disfiguring outcome on the body where it occurred (Kebede et al. 2016:565). Similarly, in Syria, appearance of the lesion followed by permanent skin mark was mentioned by most of the respondents (48

cases) and prevention of disfigurement (23 cases) was the most frequently mentioned reason for seeking treatment (Abazid et al. 2012:12).

About 257 (60.6%) of the participants in this study share the fact that environmental hygiene is important for the prevention of CL transmission. "Improved Sanitation and Hygiene" in Ethiopia is defined as "100% implementation of enhanced hygiene and sanitation when individuals demand, develop, and maintain a sanitary and healthy environment for themselves" (Federal Democratic Republic of Ethiopia Ministry of Health 2005). Ethiopia plans and implements methods to ensure that every community adopts improved (household and institutional) sanitation and hygiene, contributing to better health, a safer, cleaner environment, and the country's socio-economic growth (Federal Democratic Republic of Ethiopia Ministry of Health 2005). In the case of leishmaniasis, the burden is said to fall disproportionately on the poorest portions of the world population, where increased infection risk is mediated by poor housing conditions and poor sanitation (Alvar, Yactayo & Bern 2006:1). Environmental hygiene is also important for illness prevention because it prevents sandfly vectors from breeding near animal manure and filthy areas (Bsrat et al. 2015).

In this study, more than half of the 251 respondents (59.3%) chose indigenous medicine based on the use of herbs by traditional healers. In a comparable research of Ethiopians' treatment preferences, the vast majority [235 (59.9%)] believed that therapy from traditional healers was successful (Kebede et al. 2016:565). The use of traditional home treatments was more frequent in Colombia (Patiño-Londoño et al. 2017) and French Guiana (Odonne et al. 2011) increasing the under-recording of cases. However, the use of traditional remedies necessity rather than a choice since biomedical treatments for leishmaniasis is not available (Odonne et al. 2011). Similarly, in Ethiopia, only very few health centres diagnose leishmaniasis therefore, most cases are handled traditionally using herbal remedies or applying heat with fire from charcoal (Yohannes et al. 2019:2).

Never having a chance to learn about the disease and inadequate source of information in the locality were mentioned by 203 (47.9%) of the respondents as reasons for not being well informed about the disease. A similar study also indicates that as many as 97.3% of the participants reported that they had never had any formal education on leishmaniasis in the community (Doe et al. 2019:36). If there would be any control and prevention programmes, most of the study participants 329 (77.4%) are willing to take part as 274 (64.6%) of the participants see CL as a very serious problem in their locality. Even if awareness creation and participation of communities in control interventions are vital, Ethiopia does not have a control program against CL though there is a guideline for the diagnosis, treatment, and prevention of CL since 2013 (Yohannes et al. 2019:2). Awareness creation campaigns are also rarely available to few (1.7%) local inhabitants in West Alexandria (Randa et al. 2015: 2171). Almost all, 375 (95.7%) of the study participants in Ethiopia, did not participate in the CL control activities (Kebede et al. 2016:566). However, the availability of such control activities in the area was not mentioned in this study. So, this report may reflect the unavailability of designed control programmes rather than the low engagement of the community.

In this study, majority (64.6%, n=274) see CL as a very serious problem in their locality. Unlike this study, a smaller proportion (58.0%) of Punjab communities in Pakistan believed that leishmaniasis was a less serious disease than dengue fever, while 42.0% of respondents disagreed, arguing that because of recent severe dengue fever epidemics, people would consider CL a less serious disease (Akram et al. 2015:5). CL, on the other hand, was not regarded as a serious disease in India by any of the respondents (Nandha et al. 2013:1054). In Southern Iran (Sarkari et al. 2014:568), Nigeria (Awosan et al. 2013:261), and Paraguay (Awosan et al. 2013:261), CL was regarded a serious public health problem in 87.0%, 53.0%, and 10.0% of communities, respectively (Ruoti et al. 2013:3).

Generally, 226 (53.2%) of the participants have a favourable attitude and 197 (46.4%) have unfavourable attitudes. However, a higher proportion of the communities (> 71.6%) in Southwestern Iran, had a good attitude (Vahabi et al.

2013:865). This discrepancy could be attributable to the study participants' greater educational status in Iran, where just 6.2% were illiterate compared to 51.1% in the current study. Moderate attitude scores were reported among mothers of children infected by CL (Hejazi et al. 2010:) and in general communities in Paraguayan (Ruoti et al. 2013:3).

5.2.5. Discussion of factors affecting attitude of the community towards CL

In the bivariate analysis, age (p< 0.048 and p< 0.018), occupation (p< 0.028), place of birth (p< 0.017), and knowledge of someone infected with CL (p< 0.000) show statistical significance value for a favourable attitude of CL. As a result, those who are in the age group of 24.5-34.5 [COR=2.553; 95% CI, 1.007-6.475] and 34.5- 44.5 [COR= 3.000; 95% CI, 1.208-7.448] have a favourable attitude towards the diseases compared to those in the 18.0-24.5 age group. As people live longer, they would probably have more encounters with the disease or knowledge acquisition about the disease leading to having a better attitude towards the disease.

The multiple logistic regression analysis shows that respondents who are in the 34.5-44.5 age group [AOR= 3.584; 95% CI, 1.299-9.890] and who can read and write [AOR= 1.929; 95% CI, 1.095-3.399] have a favourable attitude than those who are in 18.0-24.5 age group and who are illiterate. Similar reports were found in a study conducted in Iran with education being a stronger predictor for the attitude (Rakhshani et al. 2017:188). Respondents with no history of knowing a person infected with the disease [AOR= 0.221; 95% CI, 0.136-0.358] are less likely to have a favourable attitude towards CL than those who previously know a person infected with CL.

5.2.6. Prevention practice of Sodo community regarding cutaneous leishmaniasis

When asked how CL was treated, the application of herbal remedies 312 (73.8%) and the use of specific drugs at health centre 187 (44.2%) were mentioned more often. Only one person (0.2%) suggested home rest without medication. Similarly,

just 48.0% of Iranians believe CL can be cured with medicine, while a lesser percentage (21.0%) believe locally accessible herbs are effective in treating the disease (Sarkari et al. 2014:568). In the case of visceral leishmaniasis, almost all study participants (97.35%) prefer going to health services first. This could be due to the fact that VL is lethal if not treated (Berhe et al. 2018:5). Moreover, half of the communities in Paraguay (57.14%) sought care from the hospital for CL/MCL (Ruoti et al. 2013:3). In French Guiana, 71.2% believed they could cure the condition at home, 65.9% would go to the hospital first, and 31.1% would utilise medicinal herbs first (Odonne et al. 2011:1232). The use of herbal medication causes underreporting of the disease making the understanding of the true burden of the disease difficult. Medical treatment at health facilities was not stated as a treatment option sought for in Ghana as 45.8% of the participants did not report the disease at the health centre because of financial constraints, ignorance and use of the herbs (Doe et al. 2019: 38).

About half 235 (55.6%) of participants in this study use preventive measures. Better preventive practices were reported in other studies. For example, the majority (79.0%, 198/250) of respondents in Paraguay believed that CL/MCL could be prevented (Ruoti et al. 2013:3). In Ecuador, many of the study participants (81.0%) had a better practice of CL. Prevention was reported (Sorocco et al. 2017:1) and more than half (62.12%) of the respondents in the Syrian Arab Republic believed the disease could be prevented (Abazid et al. 2012:10). The difference in preventive practice between participants in this study and those in Paraguay, Ecuador and the Syrian Arab Republic may be the presence of a national leishmaniasis control program since 1990 in Paraguay, and a vector control program in Ecuador. These will enhance awareness of the community hence leading to better preventive practices. In addition, notification of leishmaniasis is mandatory in these countries and passive and active detection of cases is in place (WHO, 2020). In contrast, there is no national control programme for leishmaniasis and sandfly vector in Ethiopia.

Participants in this study used bed nets 178 (42.1%), cleanliness 118 (27.9%), early detection and treatment 114 (27.0%), and insecticide 94 (22.2%) for prevention of the disease. In Iran, 16.1%, 14.8%, 3.7%, and 12.3% of the population had utilised medications, pesticide sprays, repellents, and bed nets to prevent the sickness, respectively (Vahabi et al. 2013:865). Admission to a hospital (31.2%), cleanliness (27.2%), usage of bed nets (26.0%), isolation of patients (9.2%), and dietary precaution (6.4%) were all cited as approaches for patient care with leishmaniasis in Pakistan (Akram et al. 2015:6). The use of a bed net (37.0%), insecticide (41.0%), hygiene (33.0%), adequate face and hand washing (19.0%), and bathing (16.5%) were all mentioned by participants in Southern Iran (Sarkari et al. 2014:568).

Only 35.4% of the communities in Colombia who knew the disease have practiced measures to control the disease (Pardo et al. 2006:1). The majority of physicians in Nigeria, 89.9% and 90.0%, respectively, knew vector control and treatment of sick people as approaches to prevent and control CL. However, 34.9% and 31.7% of respondents, respectively, saw avoiding close contact with an infected person and avoiding bathing in dirty water as strategies of preventing and controlling CL (Awosan et al. 2013:261). Another study conducted in Ethiopia found that participants utilised bed nets (21.7%) and DDT (19.4%) as preventative strategies (Kebede et al. 2016:566). In the Volta Region of Ghana, 26.1% of endemic communities used repellant, 17.1% slept in a bed net, 17.1% sprayed and weeded around their homes, wore protective clothing, and used herbal medication as preventive techniques against CL (Doe et al. 2019:36).

In the current study, poor prevention practice was observed on personal protective measures (wearing covering clothes 5 (1.2%), use of insect repellents 28 (6.6%) which are important as a first line defense mechanism against sandfly bite. The best way to prevent infection is to protect from sandfly bites by following these preventive measures to decrease the risk of being bitten. The use of personal protection is still considered the first line of prevention for leishmaniasis in all people working in these endemic areas because sandfly eradication is difficult. Chemoprophylaxis drugs are not effective, and vaccines are still under development (González, Solís-Soto &

Radon 2017:1). Similar reports of low use of personal protective equipment among families were seen in studies conducted elsewhere. Studies report that most of the participants did not use protective methods to protect against transmission even though they know the importance of vector control as a key to prevent vector-borne diseases (Nikookar, Pashaeei, Nikzad, Moosa-Kazemi & Davari 2015:61). The proportion of participants wearing protective clothing and using repellent (18.0%) as their ways of prevention was low in Ghana (Doe et al 2019:38). In French Guiana, methods of leishmaniasis prevention for 36.4% of the population was mainly based on the topical application of carapa seed oil mixed with annatto seed paste as repellents (Odonne et al. 2011:1232).

In this study, it was found that 251 (59.3%) use bed net when they are sleeping. With regards to the prevention of CL in the endemic communities, 39.6% of the participants reported that they always use bed net while 23.4% indicated they had never used bed net at all (Doe et al. 2019:38). The majority (85.0%) of patients with post kalaazar dermal leishmaniasis in India use bed net while sleeping (Garapat et al. 2018:5). On the contrary, out of the total 60 interviewed inhabitants in West Alexandria, only 4 (6.7%) use bed nets (Randa et al. 2015:2172). In some endemic areas, people are not using bed nets (78.0% in Afghanistan) because bed nets were too expensive (Pardo et al. 2006). Unaffordability of bed nets is an issue for only 5.8% of participants in Shahid Babaie Airbase (Saberi et al. 2012:308Bed nets provide a safe haven for those who sleep beneath them. Bed nets treated with pesticides, on the other hand, provide superior protection than untreated nets since the insecticides used to treat bed nets kill insects. The pesticides also repel vectors, which reduces the amount of insects that enter the residence and try to feed on the people' blood. The number of vectors and their lifespan will be reduced if a high level of community coverage is attained. Then, regardless of whether or not bed nets are used, community protection is guaranteed. ITNs must be used by more than half of the individuals in a community to obtain such results (Centre for Disease control 2019).

Looking into the number of bed nets in the current study, of those who use bed nets 105 (24.8%) and 69 (16.3%) have a maximum of two or three bed nets in the house, respectively. In a study conducted in Ethiopia, 203 (76.89%) of the respondents had at least one bed net in their family, and 98 (88.29%) concurred that priority was given to children in case of insufficient bed nets (Berhe et al 2018:6). On the contrary, a smaller proportion (9.6%) of communities in Pakistan use bed nets to control leishmaniasis (Akram et al. 2015:6). In Paraguay, most of the population 86.0% (397/463) never used a bed net (Routi et al. 2013:3). The malaria prevention and control programme in Ethiopia targets 100.0% household coverage with two ITNs per household in all malaria endemic areas (Federal Ministry of Health, 2020). However, there is a geographical difference in the distribution of malaria and leishmaniasis in the country. Therefore, though efforts made to prevent malaria also reduce the burden of leishmaniasis in some areas, a specifically designed national leishmaniasis control programme is still in need.

Only 19 (4.5%) of those polled have a habit of spending time or sleeping outside. Another survey conducted in southern Ethiopia found that the majority of respondents (59.2%) did not sleep outside (Kebede et al. 2016:566). Different sleeping habits were seen in the northern region of the country, where more than half of the population, 166 (62.88%), had experienced outdoor sleeping, particularly during hot weather (Berhe et al. 2018:6). The disparity is due to the temperature and humidity differences between the southern and northern parts of the country. The temperature in northern Ethiopia is hot all year, and people have a practice of sleeping outside.

In this study, more than half of the respondents (244 (57.7%) do not use a repellant. Among the Visceral Leishmaniasis prevention methods used in Western Tigray, Ethiopia, 88 (33.33%) and 163 (61.74%) respondents employ repellents and chopped plant parts, respectively, to protect oneself from biting flies (Berhe et al. 2018:6). In order to avoid insect-borne infections, the Centers for Disease Control and Prevention (CDC) suggests using insect repellents. Human breath (carbon dioxide) and skin smells attract mosquitoes and other blood-feeding insects.

Repellents are one of the personal protection measures that contain a chemical that makes the person undesirable to the vector, preventing biting. Furthermore, insect repellents are beneficial in preventing bites, skin eruptions, and rashes caused by bug bites (Anderson 2021).

In the current study area, any type of insecticide has never been sprayed in 210 (49.5%) of the participant's houses. Of those houses in which insecticide was sprayed 170 (40.0%), the purpose of the spraying was for protection against malaria. In another section of Ethiopia, the vast majority of respondents (94.50%, n=249) said they had never used insecticide spray in their house/neighborhood and those who had used insecticides. Nonetheless, during the rainy season, they conduct it secretly on mosquito breeding spots around their compound (Berhe et al. 2018:6). The Ethiopian Ministry of Health plans a national malaria control program that will use indoor residual spraying to reach more than 85.0% of residents living in epidemic areas (Federal Ministry of Health 2020). Even though this is for malaria prevention, the low coverage of indoor residual spraying shown in these studies conform insufficient implementation of the programme.

When asked about their preferred time of work at peak temperature, the respondents prefer to work at daytime 148 (35.0%) and both at daytime and night (39.0%). Another study conducted in Ethiopia found that the majority of the study participants worked on their farms during the day [375 (95.7%)] (Kebede et al. 2016:566). Since a sandfly bite at night, working in these times pose exposure to the bite and disease transmission.

Two hundred and ten percent (49.4%) of the respondent have good prevention practice while 214 (50.4%) of the respondents have poor practice. According to an Ethiopian survey on visceral leishmaniasis, more than half of the respondents (68.6%) followed proper kala-azar prevention and control procedures (Alemu, Alemu, Esmael, et al. 2013:1). People tend to have stronger preventive procedures against visceral leishmaniasis due to its deadly nature. In Iran, 32.5% of women with afflicted children performed poorly, 31.3% performed averagely, and 36.1% of

moms with affected children performed exceptionally (Hejazi et al. 2010:36). In Iran, however, participants' practice scores were lower among communal families (with a mean score of 5.6 out of 36 questions linked to leishmaniasis practice) (Rakhshani et al. 2017:189). Low practice scores suggested that health researchers and policymakers would need to implement practice change measures to help improve the practice. Understanding people's perspectives and practices is critical to the successful implementation of CL control actions in CL-endemic areas (Rakhshani et al. 2017:190).

5.2.7. Factors affecting practice against cutaneous leishmaniasis

In multiple logistic regression, only knowing someone infected with the disease shows statistical significance relation with good practice regarding CL [OR=0.315; 95% CI, 0.195- 0.510]. Similarly, a family history of CL was a stronger predictor for the practice in Iran (Rakhshani et al. 2017:188).

5.2.8. Discussion of overall knowledge attitude and practice

Regarding the overall Knowledge of the Sodo community regarding CL, 263 (61.9%) have satisfactory knowledge of CL. Similar reports were seen from the southern part of Ethiopia where (67.6%, n=265) study participants were knowledgeable (Kebede et al. 2016:565). In west Alexandria, Egypt, among the 20 primary health care physicians, 55% of the physicians had satisfactory knowledge (Randa et al. 2015:2177). In Colombia, only 15% of homeowners had a "very excellent" understanding of CL, compared to 56.3% who had a "bad" or "poor" understanding of CL, according to these research (Pardo et al. 2006:174). A survey conducted in Iran also shown that people in Iran had a poor understanding of CL (Rakhshani et al 2017:190). According to another study conducted in Iran, 211 people (52.1%) were unaware about CL (Vahabi et al. 2013:866). The discrepancy in the community awareness report in the aforementioned research can be explained by the study population and study period differences.

In the current study, the percentages of study participants showing favourable and unfavourable attitudes about CL are 53.4% and 46.6 % respectively. A similar study

in Ethiopia revealed a lower percentage of positive attitude among 162 (41.3%) (Kebede et al. 2016:566). A higher percentage (85%) of a good attitude score was seen among Egypt health care physicians showed a good attitude about leishmaniasis (Randa et al. 2015:2177). Health care physicians are would probably have better disease awareness owing to educational preparation and clinical experience. In Iran, average scores of attitudes were reported among households (Rakhshani et al. 2017:190) and mothers of children with CL (Hejazi et al. 2010:37). About half 213 (50.4%) of the participants in this study show good preventive practice against the disease while the rest 210 (49.6%) have a poor practice of CL. Mothers of children with CL have better practice compared to participants in this study as only 32.5% of the mothers had weak performance (Hejazi et al. 2010:37). This may be because females have a better attitude than males as shown previously in this study (Section 4.3.3.2). However, the practice score of participants in this study is higher than reports from southern Ethiopia by Kebede et al. (2016:565) where 62.5% of the population has poor practice. This may be because of differences in the study period. With the current study conducted recently, communities may have better access to knowledge. Students attending Isfahan's Shahid Babaie Airbase's middle and high schools were likewise found to have a lack of CL practice (Saberi et al. 2012:308). This may be owing to the age factor as students in school are younger than populations in randomly selected communities.

Comparing overall practice with knowledge and attitude in the current study, the result indicates of 262 (61.9%) respondents with satisfactory knowledge 173 (40.9%) have a good practice. The association between knowledge and practice was statistically significant at $p < 0.000$ accordingly the odds of having a good practice are 5 times higher for those who have satisfactory knowledge than those who have unsatisfactory knowledge. A similar relationship was seen between attitude and practice as of 226 (53.4%) participants who have favourable attitude, 150 (35.5%) also have a good practice. Individuals with a positive attitude were three times more likely to have a good practice, which was statistically significant at $P < 0.000$. Both knowledge and attitude had a strong connection with practice (p-value 0.001) in a KAP study of Iranian homes (Rakhshani et al. 2017:188). This

indicates the effectiveness of educational activities in endemic areas for disease prevention. Indian houses that were exposed to behavioural change activities that largely utilised group and interpersonal communication had a higher KAP than those who were not, including acceptance of indoor residual spraying (IRS) as a preventive strategy. As a result, BCC initiatives are a critical component of VL elimination strategies (Srinivasan, Ahmad, Raghavan, Kaushik & Pathak 2018:1). People's health information behaviors pertain to how they seek, access, analyse, categorise, and use relevant health-related information to accomplish desired health behaviors. Health information behaviors are inextricably linked to appropriate and consistent performance of these behaviors. Practice efforts to seek and get health-related knowledge are linked to more health-oriented thinking and, in general, healthier behaviors (Stefan 2015:737).

5.3. DISCUSSION OF PHASE TWO – FACTORS AFFECTING CUTANEOUS LEISHMANIASIS

Different factors affect the transmission of CL. Identifying these factors can help design preventive measures. Here demographic, household, host, environmental, and animal factors that are determining the disease transmission in the area are discussed in comparison to similar studies.

5.3.1. Discussion of residential features

Of those respondents who have a farm or whose livelihood depends on the farm, the farm location is near home (29.3%), both near home and near a river (17.7%) and a river (3.17%). This shows that half 50.0% (681/1356) of the respondents have or work on a farm which is found near home or a river.

The presence of a farm near a river has many implications linked to CL. A farm found around a river is likely to be involving irrigation thus, farmers who irrigate early morning or overnight to avoid the hottest periods of the day, are exposed to bites from nocturnal sand flies. Furthermore, the expansion of intense farming activities surrounding a supply of water is usually accompanied by the rise of sheep and cattle pastoral husbandry. Animal dung is stored and used on a large-scale

farm to offer ideal conditions for sandflies to develop because shelter and a microclimate favorable to their activities are generated (Nouiri, Chemak, Mansour, et al. 2015:94).

5.3.2. Household factors

People who live in the traditional mud and grass huts known as "Gojo bet" may be at danger of contracting the disease, as studies have linked mud houses to an increased risk of leishmaniasis. Houses constructed primarily of mud in Asia are linked to VL (Bern et al. 2010:2). In a leishmaniasis-endemic district of Nepal, 85% to 87% of the dwellings were made of mud and dung poured over a stick framework (Bern et al. 2000:186).

Looking into the wall condition, cracks or holes were not observed in majority of the houses 289 (76.3%). Wall conditions in nearly one fourth 90 (23.7%) of the households represents the presence of risky wall condition (holes formed (1.3%), cracked (10.6%) and with both crack and holes (11.8%) creating favourable vector hiding places in the house which in turn will increase exposure of household members to vector bite and consequently disease transmission. A study conducted in Nepal for the identification of leishmaniasis risk factors indicates that cracks in the walls of the house posing risky conditions (Bern et al. 2000:186).

The roof was made of steel in the majority (70.1%, n=266) of the houses in the study area. Houses having this type of roof are less risky to disease than houses with a thatched roof made of grass or straw. In a case-control study conducted in Nepal Case households were more likely to have thatched roofs (68.0% compared with 54.0% among control houses; OR _ 1.8, 95% CI 0.9–3.4) (Bern et al. 2000:186). Floor-type of houses in the study area were mostly (97.0%) made of mud and out of these, 91.0% were plastered with cow dung. Mud floors with the addition of cow dung on top of it create a perfect microclimate for the sandfly therefore increasing risk. In Nepal, humidity in the floor of the house was related with higher risk of the disease (Bern et al. 2000:186).

When health extension workers visited the residences, 355 (93.7%) had a working latrine, and the latrines were in fair condition. Open defecation is mostly a rural phenomena most prevalent in Ethiopia, resulting in organic matter accumulating on the ground. This could provide a breeding ground for sandflies, increasing the risk of sickness. However, studies in southern Ethiopia found no link between CL and the presence or absence of a toilet, a waste disposal facility near the house, or a waste disposal pit near the house (Negera et al. 2008:886).

Only 88 (23.2%) of the dwellings had screens on the windows or doors. Leishmaniasis risk may be reduced as a result of reduced vector exposure. Leishmaniasis, malaria, and other mosquito-borne infectious diseases can be reduced by screening windows and doors in the home. To lower the risk of leishmaniasis, control measures focusing on preventing vector contact and implementing short-term treatment schemes are recommended (Reithinger, Mohsen & Leslie 2010:2).

5.3.3. **Environmental factors**

Half (52.95%) and 21.98% of the household have acacia and sensel tree around the house, respectively. Studies in Ethiopia indicated that the Leishmania vector is associated with red acacia, balanites trees and termite hills (Assefa 2018:2). These trees serve as a resting place for the sandfly and may increase the exposure of individuals to sandfly bite.

Nearly a quarter (23.23%) of the respondents reside in a place near to the presence of a backyard and a field. Only 13.94% of residents say that they have a public assembly area near their home. Majority (72.94%) of the respondents in the households confirm the existence of a farm within 300 radios. Sand flies are not strong flyers, and they normally stay within 300 meters of their nesting place (Negera et al. 2008). In a study conducted in Ethiopia to identify associated factors of leishmaniasis among clinically suspected patients attending hospital, the proportion of respondents having farm around their house is lower (38.04%)

(Bisetegn et al. 2020:6). But a report from the North part of Ethiopia indicates that majority (80.2%) of the study participants live in a house near the farm (Bsrat et al. 2015:6). The presence of a farm creates a suitable condition for sandfly and expose farmers or individuals living near the farm to sandfly during farm work. The presence of cave or gorge in this study area was 48.60%, 38.79% and 23.75% in villages, in working area and around household area, respectively. Greater proportion of houses (62.2%) in leishmaniasis endemic areas of Ethiopia are found to be located near the cave or gorge (Bsrat et al. 2015:6).

Water sources for individuals living in the study area are tap water (39.0%), river (28.0%), hand-dug well (23.0%), and spring (10.0%). Adding the three water sources except tap water makes 61.0% of the contribution. This means that majority of the individuals live near a water source or are in close contact with these. Leishmaniasis is more likely to impact the poorest people and marginalised groups, particularly those that live near water resources, dwell in damp houses, and are near garbage, sewerage, and livestock farms (Oryan & Akbari 2016:4).

5.3.4. Host factors

Under this section, host factors which may contribute to the occurrence, transmission, and prevention of the diseases are presented. Majority (70.28%) households keep dry wood stored inside the house which might give access to the creation of termite hills under the woods and the inner side of termite mounds can be considered as chief outdoor diurnal resting places for sand flies (Wijerathna et al. 2020:2). The presence of termite hills differed considerably between patients and controls in case-control research in Kurunegala District, Sri Lanka, to evaluate socioeconomic, demographic, and environment characteristics related with CL. It was linked to an increased risk of infection, with the odds of contracting the disease increasing by 2.4 times (Wijerathna et al. 2020:8).

Households having a history of travel made up 12.39% of the total. Adventure travel and outdoor sports are becoming increasingly popular among international travelers, putting them at risk of developing leishmaniasis (Pavli & Maltezou 2010:

e1033). This type of infection is called imported leishmaniasis (Pavli & Maltezou 2010: e1033) and is also common in non-international travellers who visited an area with higher endemicity. Because non-immune people were introduced into areas with existing transmission cycles, people who came from the highlands of Tigray and Amhara regions of Ethiopia for weeding and harvesting sesame were at higher risk of leishmaniasis infection than those who lived permanently in and around Humera (endemic area for VL) (Tedla, Bariagabr & Abreha 2018:7). When these individuals return to their original location, they may act as a reservoir for the host and a source of infection for others. The introduction of the parasite by migrant agricultural labourers returning to their communities after completing seasonal work on the country's border is one probable reason for an epidemic that occurred in a previously non-endemic area of Ethiopia (Libo Kemkem) (Sordo, Gadisa, Custodio, et al. 2012:956).

The use of insecticide-treated bed nets was experienced by 40.86% of the participants in the study. When asked about the sleeping condition, almost equal percentage of individuals said that they sleep on mat/matters with bed net (47.12%) and mat/mattress without a bed net (46.24%). A bed net is a net that hangs from the ceiling above a sleeping space, usually a bed or matt, and provides a physical barrier between vectors and the individual who is at risk of contracting the disease. More importantly, an insecticide-treated bed net gives an added benefit by eliminating vectors that do get through the net before they reach the sleeping person and protecting the person sleeping beneath the net, even if the net has small holes in it (CDC 2020).

According to majority of the participants (87.88%), the village is not expanding to the previously abandoned forest. Expansion of villages to previously abounded forest cause risk to vector-borne disease as it brings reservoir hosts and vectors closer to the human population. It has been described that deforestation has led to an increase in leishmaniasis it makes the sandfly vectors become peri domestic (Oryan & Akbari 2016:4). When forests are removed, new vector breeding sites are produced, and the ecology is affected by changing borders. By redesigning current

ecosystem borders, which might serve as points of contact between humans and forest pathogens, forest removal changes ecosystem dynamics and creates new breeding grounds for disease vectors. People who live in or near forests are at a substantially higher risk of illness due to increased vector contact (Gottwalt 2013:1).

In the current study, of the 651 (48.0%) individuals who have a habit of sitting in the home yard at dawn/dusk, 25.34% do this always, 41.01% frequently and the rest 20.89% sometimes sit in the home yard at dawn/dusk. Sitting outside at dawn or dusk exposes people to female sandfly bites, which require protein to lay eggs. The majority of sandfly species eat around dusk and at night, when the temperature drops and the humidity rises. Although daylight biting can occur indoors in gloomy areas or among shaded vegetation/trees, especially if disturbed by human activity, it is more likely to occur at night (European Centre for Disease Prevention and Control 2020).

5.3.5. Animal factors affecting CL

Different researchers indicate an association between CL and the presence of animals around the house. Therefore, in this section details of animal factors and their distribution among the households will be described. Hyraxes exist in working areas (47.20%), in villages (44.10%), and near the house (31.49%) and a half (50.07%) of the respondents verify the presence of hyrax burrow within 300meter radius of the house. Hyraxes, Procavia capensis and H. brucei, have been identified as reservoir hosts of *L. aethiopica*, with natural infection rates ranging from 21 to 27%. *Phlebotomus longipes* and *Phlebotomus pedifer*, two known CL sandfly vectors in Ethiopia, readily feed on hyraxes and share their environment. Sandflies find a good nesting environment in the organic debris deposited in their hyrax's latrines (Lemma 2018:314).

Only rock hyraxes were discovered to be Leishmania parasite reservoirs in Ethiopia. Natural Leishmania infection in bats has been documented in Ethiopia, and the data suggest that bats may play a role in the transmission cycle epidemiologically. Bats

and sand flies share a lot of living space because they're both cave dwellers with a lot of opportunity for sand flies to feed on bats (Kassahun et al. 2015: 166).

The bulk of the respondents in this study (80.53%) have domestic animals. The majority of the time, these domestic animals are kept inside the house (29.3%) or in a stall close to the house (61.44%). Four hundred and seven people (30.01%) indicated they have a tendency of depositing animal manure near their homes. Leishmaniasis is commonly seen in agricultural settlements in the South, where cattle and other livestock are maintained near to human houses (Bern et al. 2010:2).

5.3.6. Demographic characteristics and residential futures on CL occurrence

From the secondary data collected from the district health office, Kola Nurena and Beki are kebeles with highland topography and are not considered for the application of malaria prevention strategies. Reports of rare cases of malaria in these kebeles are associated with people having a travel history to other malaria-endemic regions. In Ethiopia, vector control program for leishmaniasis is not available but the distribution of bed net and spraying insecticides is done to prevent the disease following steps in malaria control (Deribe, Meribo, Gebre, et al. 2012:8).

5.3.7. Association between household factors and cutaneous leishmaniasis

In Afghanistan application of screens on window was found to be a significant determinant of disease occurrence in the households. Houses which have screens were negatively associated with CL status (Reithinger, Mohsen & Leslie 2010:2). Screened windows that allowed light to enter, humidity to exit and at the same time prevented sandflies from entering have demonstrated a protective factor against leishmaniasis infection (Calderon-Anyosa, Galvez-Petzoldt, Garcia & Carcamo 2018:1551).

5.3.8. Environmental factors and cutaneous leishmaniasis

Environmental factors selected to be affecting CL presence are the existence of farm in 300m radius of the house, presence of acacia and sensel trees, and

presence of cave or gorge near home, near the village and working area. The result of the bivariate and multivariate analysis shows a significant relationship between the presence of sensel tree and cave or gorge near home with disease occurrence. This result shows that individuals living near sensel tree are at double risk of getting the disease [AOR=2.31; 95% CI, 1.507-3.555]. Moreover, the presence of a cave or gorge near home increases the likelihood of infection by seven-fold [AOR=7.02; 95% CI, 4.419-11.273]. The presence of trees that can provide refuge for hyraxes is crucial to the spread and development of leishmaniasis. Because these plants provide protection against predators and vector proliferation in regions where these plants are abundant, these trees may also act as a breeding/resting site for vectors. Adults repose in humid areas during the day, such as inside human houses, tree fissures, and earth cracks (Elnaiem, Khogali, Alsharif, et al. 2020:2). In Sudan, displaced persons living in acacia-rich areas saw one of the greatest epidemics of visceral leishmaniasis (Lima, Mesquita, Skrip et al. 2016:1).

Participants residing near caves/holes were shown to be 46.0% (OR = 1.46; 95% CI: 1.12-1.90) more at risk than those who lived far away from caves/holes, animal burrows, animal excrement, and farmland, according to a study done in Ethiopia's northern region (Bsrat et al. 2015:6). Because of the strong relationship between sandflies and water, these caves/gorges are likely to be hyrax habitats. Because sandflies are short flyers and stay in their breeding or resting places, the distance of the reservoir host has a significant impact on the likelihood of humans getting bitten by them (Bsrat et al. 2015:7).

5.3.9. Host related factors and their relation to cutaneous leishmaniasis

No history of travel was shown to be substantially linked with CL, indicating that the disease is endemic in the study location. Travel history was found to be related with CL, contrary to the findings of this study. Similarly, a study on CL in Ethiopia found that travel history was substantially linked with CL (AOR 13.9, 95% CI 4.4–14.3, $p < 0.01$) (Eshetu & Mamo 2020:8). In a comparable study conducted in Ethiopia, where labor migrants from the highlands to the agricultural fields had greater infection rates than those who resided continuously in and near the endemic area, the origin

of the place was also substantially linked ($p < 0.05$) (Tedla et al. 2018:1). The majority of individuals admitted to a hospital in England (71.0%) had previously been to endemic areas (Wall, Watson, Armstrong, Chiodini & Lockwood 2012:1). The differences in this report are due to differences in CL endemicity in the research location. People having a history of travel to endemic areas have been linked to disease occurrence in non-endemic locations, but as all residents in endemic areas are equally at risk, travel history is not a distinguishing feature of disease status in endemic areas.

Sleeping conditions like sleeping in the home yard or compound and covering face and hand while sleeping were significantly associated with this study. Similar findings were recorded from Western Tigray (Tedla et al. 2018:1), Northcentral (Eshetu & Mamo 2020:8), and Northeast (Bisetegn et al. 2020:8) Ethiopia. Working or sleeping outdoor unprotected from sandfly bites at night during agriculture seasons or guarding cattle are the main reasons for the VL incidence (Lemma 2018:317).

In this study, other behavioural activities like spending time near a gorge/cave and irrigation at night were found to be significantly contributing factors for CL occurrence. In Ethiopia, hyraxes live mostly in crevices of basalt rocks in the gorges and tree cavities in forests (Lemma 2018:317). Individuals who spent time near the gorge/cave are prone to sandfly bites. A study in Ethiopia indicates that the presence of caves and gorge in the vicinity were highly associated with CL. As a result, individuals near caves/gorge three times higher risk (Yohannes et al. 2019:8).

Same to the findings in this study, the use of personal protective measures was found to be effective in Colombia (Rojas 2001).

5.3.10. Animal-related factors affecting cutaneous leishmaniasis occurrence

Individuals living with hyrax around the house [AOR=2.49; 95% CI, 1.679-3.708] and have a habit of dumping cow dung near the house [AOR=2.35; 95% CI, 1.549-3.557] are twice at risk, in comparison to others with out hyrax near by or do not dump cow dung near their house.

Hyraxes, which provide an alternate diet for sandfly vectors, mediate permanent CL foci in Ethiopia (Yohannes et al. 2019:2). Hyrax was shown to be linked with CL in the Tigray region of northern Ethiopia (OR = 4.15; 95% CI: 2.64–6.53), confirming the findings of the current investigation (Yohannes et al. 2019:8). Similarly, after controlling for other risk variables, the presence of hyrax in the village and the existence of CL lesion demonstrated a statistically significant relationship with the incidence of CL in northeast Ethiopia (Bisetegn et al 2020:7). Participants in north Ethiopia who resided near a hyrax shelter had a two-fold increased risk (OR = 2.54; 95% CI: 1.87-3.46) (Bsrat et al. 2015:5). Hyraxes are the only reservoir hosts of CL in the Ethiopian and Kenyan highlands, where they share a habitat with Phelobotomus species. CL epidemics in the Ethiopian highlands are frequently linked to the presence of hyraxes (Lemma 2018:314).

Individuals who dump cow dung near their homes were shown to have twice the risk of contracting the virus in the current study. Domestic animals, primarily cattle, in close proximity to houses have been highlighted as a risk factor for leishmaniasis (Oryan & Akbari 2016:4).

5.3.11. Discussion of association of all factors

Living in Kela Zuria kebele [AOR=0.01; 95% CI, 0.003- 0.049], Michael Semero [AOR=0.07; 95% CI, 0.020-0.203] and Genete Mariam [AOR=0.16; 95% CI, 0.046-0.506], use of personal protective measure [AOR=0.14; 95% CI, 0.051- 0.362], sleeping hand/face covered [AOR=0.05; 95% CI, 0.024-0.124] and presence of screen on window door [AOR=0.088; 95% CI, 0.035-0.214] were protective factors. Living in Kela Zuria, Micheal Semero and Genete Mariam reduce the log odds of infection by 4.2, 2.7, and 1.8, respectively. Additionally, the use of personal

protective measures, sleeping hand/face covered, and having a screen on window/door reduce the log odds of CL by 1.9, 2.8, and 2.4. Kela Zuria is the kebele containing the administrative center of the district called Buee and is characterized by an urban setting compared to other Kebeles. Though CL is observed in both rural and urban areas, people in an urban setting may have better preventive practices due to better access to health information. Plus, environmental factors (Presence of cave/gorge, trees...) favoring the survival of hyraxes and sand flies are prevalent in rural settings compared to urban. However, urban settings also have a factor to contribute to disease transmission. One example may be an outbreak that has occurred in Ethiopia in Siliti district because of the construction of a bridge which creates a suitable microclimate for sandfly to reproduce underneath *(*Negera et al. 2008).

5.3.12. Discussion of multiple correspondence analysis and hierarchal clustering

Adjusting for other variables, kebele, a screen on window or door, presence of Sensel tree and cave/gorge near home, different host-related factors (such as travel history, sleeping at the home yard, irrigating crops at night, spending time near cave or gorge, use of personal protective measures and sleeping hand/face covered), presence of hyrax near home and in the village and dumping animal dung near home were significantly related with CL in logistic regression. The multiple correspondence analysis followed by hierarchical clustering help understanding the overall relationship between selected factors and classify individuals into distinguished groups. If put in order of importance or significance, the following were observed from the variable category- MCA map comparing variable proximity to CL occurrence

1st Cave/gorge near home.

2nd Dumping animal dung near home.

3rd Presence of sensel tree.

4th Presence of hyrax near home and irrigating crops at night.

5th Use of personal protective measures.

6th Sleeping hand/face covered and sleeping at home yard.

7th Spending time near cave or gorge and

8th Screen on window or door.

Consequently, investments can be made on awareness creation of the community to avoid dumping animal dung near home, stop irrigating crops at night, use personal protective measures, sleep hand/face covered, forbid sleeping at the home yard, and put screen on window or door. In addition, environmental management such as insecticide spraying of cave/gorges, sensel trees, and other hyrax borrows and sandfly resting places near residential areas could prevent the community from getting infected.

5.4. DISCUSSION OF PHASE THREE – PREVALENCE AND SPECIES IDENTIFICATION SURVEY RESULTS

This section presents discussions on the current CL prevalence rate and compares the finding to similar studies. Disease characteristics observed are illustrated with an explanation of the clinical presentation of the disease.

5.4.1. Prevalence and Disease Characteristics of cutaneous leishmaniasis

Out of the 379 houses visited 110 houses (29.0%) were harbouring at least one person positive for CL (Past or active). From 1,356 persons investigated, 123 (9.07%) were identified with suspected cases of CL. Of these, 49 (3.61%) were with active CL infection during the visit, and 74 (5.45%) were infected in the past (With scar).

The entire percentage (active and scar) in this study was 9.07%, however a study investigating the status of CL in Ochollo found a substantially higher percentage. Thus, the overall prevalence of CL, comprising both scar and active lesion, was 65.8% in 523 school children, with 313 (59.8%) having scars and 21 (4.01%) having active lesions, while 8 (1.5%) of the cases had both scars and active lesions (Bugssa et al. 2014:111). Ochollo is an endemic focus for leishmaniasis and has been reported to be one of the hotspots of CL in Ethiopia since 1987

(Mengistu, Laskay, Gemetchu, et al. 1992:149). Hence, the presence of individuals with scar formation from past infection is justified. However, the percentage of reports of the active lesion is slightly higher (4.01%) than the findings in the current study (3.61%).

Another study conducted to investigate an outbreak of CL in Silti District south Ethiopia area, not known to be endemic for the disease before, reported 4.8% (92/1907) total prevalence of active CL (Negera et al. 2008:885). This outbreak investigation suggests that the construction of a bridge and creation of favourable vector reproduction site underneath was the cause of the outbreak. Another study in the capital city, Addis Ababa observed 35 cases of CL (total sample size not available) with nine active and 26 healed skin lesions (Lemma, Erenso, Gadisa, et al. 2009:4). In recently conducted study in Northern Ethiopia, the overall prevalence of CL was found to be higher than the report in the current study with 14.0% (6.7% for active lesion and 7.3% for a scar) (Bsrat, et al. 2015:3). This report presents a higher prevalence rate in Ethiopia. However, another recently conducted study in the same region (Tigray North Ethiopia) indicates that 2.3% of the population have active CL and 20.9% present scar (Yohannes et al. 2019:6). The later study surveyed a total of 9622 inhabitants in 1721 households in different sub-districts covering a wider geographical area in the region. The reasons for variation as described by the author of this study were heterogeneity to exposure (entomological, environmental, or behavioural factors) and micro-ecological variation (Bisrat et al. 2015:7).

Two hospital-based studies in Addis Ababa (Bekele et al. 2014:6) and North Central Ethiopia (Eshetu & Mamo 2020:3) reported a prevalence of 234/1651 (14.2%) from 2007 until 2010 and 888/58,163 (1.53%) from 2012 until 2017 respectively. Both studies show evidence of an increasing trend of CL with 12.8% in 2007 to 42.7% in 2010 and 0.9% in 2012 to 3.5% in 2018 respectively for a study conducted in Addis Ababa and north central Ethiopia. The populations in these studies are with suspected cases of CL since these are hospital-based studies. The higher percentage of confirmed cases in Addis Ababa is because infected people tend to choose ALERT Hospital established in 1932 because it has long been known for

treating dermatological problems. However, a much higher percentage of CL positivity 50.56% (179/354) was recorded from questionnaire survey administered to patients visiting the dermatology department of the referral hospital in Desse Town (North Central Ethiopia) in 2018 (Eshetu & Mamo 2020:3). People previously used traditional pharmaceutical alternatives, and those that visit health facilities prefer to go to other hospitals where leishmaniasis treatment is widely available, according to the author. Since the hospital where the questionnaire was administered began servicing CL patients, a large number of people have traveled from far and wide to seek CL treatment there. As a result, the increased prevalence of CL cases in 2017 and 2018 supports this theory.

Despite the fact that there is no statewide prevalence of CL in Ethiopia, community-based cross-sectional studies and hospital-based surveys show significant variation. The overall random pooled prevalence of leishmaniasis in Ethiopia was 19.0%, according to a systematic literature review and meta-analysis study (95% CI 14 – 24%). The presence of a strikingly high between-study variability with the diagnosis approach contributing to the heterogeneity of studies was also highlighted in this investigation (Assefa 2018:1).

In general, a risk map design research for CL in Ethiopia based on environmental factors found that 28,955,035 Ethiopians are at risk for CL out of a total population of 86,613,986. (Seid et al. 2014:377). To put it another way, one-third of Ethiopia's population is at danger of contracting CL.

More than half (59.18%, 29/49) of people with the active infection get it treated and of those treated, 72.41% of patients with active cases use herbal medicines. Of the 62 (83.78%) of the participants with past infection who seek treatment, majority (82.08%) visited herbal healers and only seven patients (11.29%) go to modern health facilities for treatment. When asked who in the family was infected in the past, 47 (63.5%) indicated as self, and 27 (36.48%) said that it was another member of the family. No drugs for leishmaniasis are registered in Ethiopia and the few drugs currently used are not easily accessible to the Sodo community as

treatment services are provided in few health facilities. To get modern medication, one must travel to the capital city Addis Ababa and be admitted to ALERT hospital for days and months. This is not the option mostly entertained by rural communities as they are busy working on the farms and since CL is not fatal, they mostly choose to get it treated by locally available traditional medications. Traditional medicine is used by around 80.0% of Ethiopians and 90.0% of livestock for health treatment, and more than 95.0% of traditional medicine preparations are made from plant sources (Fenetahun, Eshetu, Worku & Abdella 2017:85).

The shortage of current medications for leishmaniasis treatment necessitates the hunt for novel antileishmanials. Available treatments might be highly toxic, require long-term use, and induce significant adverse responses, leading to therapy discontinuation and failure. Furthermore, antileshmanial medicines have only species-specific activity, and their use does not guarantee full parasite removal (Menezes et al. 2015:7). The comprehensive literature review study on medicinal plants used by Ethiopians to treat leishmaniasis reveals that alternative drug formulation is urgently needed. Natural product screening for potential application in leishmaniasis treatment may be considered (Aschale, Wubetu & Reta 2018:1). Traditional healers use roughly 28 plant species to treat leishmaniasis, according to this review. These drugs were used to treat 31.0% and 24.2% of CL and visceral leishmaniasis, respectively (Aschale, Wubetu & Reta 2018:1).

5.4.2. Socio-demographic features of persons with cutaneous leishmaniasis

Looking into the kebele in which positive cases belong, 34 (34.69%) were from Kola Nurena kebele. This may be because the kebele is with favourable conditions for disease transmission as 27 (79.4%) and 24 (70.5%) out of 34 positive cases in this kebele testify that there is hyrax and cave/gorge near home. In addition to this, Kola Nurena is a highland area and is excluded from the application of malaria control programmes.

Females were with a slightly higher percentage of suspected CL (9.49%) than males (8.6%) in this study. This may be because females in this group are less

educated than males as 43 (65.0%) out of the 66 clinically positive females are either illiterates or are only able to read and write. This percentage in the case of males 24 (42.0%) out of 57 clinically positive being illiterate or able to read and write. The chi-square test revealed that the difference in educational status was statistically significant (X2=11.387, p 0.02255). Similarly, a study found no difference in the frequency of CL cases between men and women, with 47.8% (44/92) males and 52.2% (48/92) females (Negera et al. 2008:885). CL patients with active and healed lesions were 17 (48.6%) males and 18 (51.4%) females, according to Lemma et al. (2009:4). Males (7.6%; 74/979) had greater CL proportions than females (5.9%; 67/1,127) in northern Ethiopia (Bisrat et al. 2015:4) and north central Ethiopia (Eshetu & Mamo 2020:5).

Other studies have found that men had a higher incidence than women, owing to the fact that men work or sleep in crowded and infected areas and are thus more exposed to infected vectors than women (Oryan & Akbari 2016:5). Males may have closer contact with the sandfly's environment through occupation, leisure, and cultural activities (such as farming, rearing cattle, staying around gorges, and/or farmland for a long period of time), according to the authors (Eshetu & Mamo 2020:6).

In the current study individuals who are illiterate (10.35%) and who are only able to read and write (10.81%) are the ones holding a bigger proportion of the suspected cases. Like this, a study in Silti district, Southern Ethiopia noted that illiterate participants were significantly linked to CL with p- value < 0.001 (Negera et al. 2008:887). A hospital-based study in north central Ethiopia shows that 65.4% and 53.9% of respondents positive for CL, received no formal education and are attending secondary education respectively (Eshetu & Mamo 2020:6). There is a well-known, significant, and consistent link between education and health that has been found across numerous nations and times, as well as across a wide range of health indicators. The disparities between the more and less educated are important, as higher levels of education lead to distinct ways of thinking and making decisions (Cutler & Lleras-Muney 2008).

In the current study, the proportion of CL positivity was slightly higher (9.85%) in respondents whose occupation involves farming than those who have a non-farm related job (8.39%). Similarly, in north central Ethiopia, a higher percentage (68.5%) of positivity was seen among farmers compared to participants with other jobs which was statistically significant in univariable logistic regression [COR= 4.5; 95% CI, 1.9–10.1] (Eshetu & Mamo 2020:6). Farmers may be more susceptible to CL than non-farmers since they are more exposed to sandfly bites in the farm field and irrigation areas (Eshetu & Mamo 2020:7).

Considering the age of participants in the current study, out of the 123 with CL, ten were between 5-10 years, and all (100.0%) of them were positive (eight with active CL and two with scars). CL is predominantly present in younger Ethiopian populations between zero to ten years old (Yohannes et al. 2019:9). For example, a study conducted in northern Ethiopia indicated that 0-9 years were highly affected group (Yohannes et al. 2019:9). Another study conducted among primary school children in southern Ethiopia shows that 6-10 years are the ones carrying a higher proportion of active infection (Bugssa et al. 2014:114). Due to lack of mature immunity, risk to CL increase with age but after 15 years the individuals become immuned. In Turkey, many CL cases are seen in childhood, and children 6-10 years (39.43%) are at greater risk than adults. This may be because children do not attend to protective measures against sandfly such as covering hand and face while sleeping and use of personal protective measures in addition to playing outside at dusk. Adults in endemic parts of Ethiopia are also thought to be less likely to have active lesions because they have already recovered from a childhood Leishmania infection, making them resistant to the development of clinical infection (Pareyn, Kochora, Rooy, et al. 2020:2).

Flowing 5-10 years, 25-32 and 35-44 years were found with CL. Other studies in the country reported differently with the most affected group of age being 11-20 years (46.7%) followed by 0-10 years (33.7%) (Negera et al. 2008:885) and 10-19 years (31.4%) (Lemma et al. 2009:4). Similarly, 10-19 years (Bisrat et al. 2014:3) and 11-

20 years followed by 21-30 years were identified as the most affected population (Bekele et al. 2014:6). While it is difficult to draw generalisation from these studies because of the difference in the age categories, it can be noticed that younger individuals between 10-30 years are most at-risk groups. The reasons for the population in this group being at high risk was outdoor activities performed by individuals engaging in farm activities, keeping crops from wild animals, the habit of going to church early morning which are located near to escarpments.

5.4.3. Type of cutaneous leishmaniasis and body parts affected by active lesion

Among the active CL, cases confirmed clinically, the majority 87.79% (43/49) were presented with LCL and the rest 6 (12.24%) were found to have MCL. In north Ethiopia, LCL was the only clinical presentation among patients (Bsrat et al. 2015:3). Out of 888 patients in north central Ethiopia, 792 (89.2%) had LCL, 35 (3.9%) had DCL, and 61 (6.7%) had MCL (Eshetu & Mamo 2020:4). LCL is the most common clinical manifestation of leishmaniasis, characterized by itchy sores and lymph node inflammation on the arms, legs, or face. The sores acquire a red elevated edge and a depression in the middle over time, which may heal or harm surrounding tissues (Nuako 2016: 26).

In the present study, most of the patients 41 (83.67%) had single lesions and only three (6.1%) developed three or more lesions. Similar study revealed that most of the patients in south Ethiopia, 46.7% (43/92) had single lesions and only 17.4% (16/92) developed three or more (Negera et al. 2008:885). As cited by Bsrat et al. (2015:4) number of lesions in 86.10% was one and the remaining 15.8% presented two or more lesions. The majority of respondents in Sri Lanka (89.8%) (Wijesinghe, Gunathilaka, Semege, et al. 2020:1) and in Turkey (65.85) (Serarslan, Ekiz, Ozer & Sarikaya 2019:113) presented with a single lesion.

The duration of the lesion in this study was determined by interviewing the patients and most of them, 20 (40.8%), had a lesion that had developed between seven months and one year ago. With 16 (32.65%), the lesion started in less than seven

months. From patients who seek health care from 2007 to 2010 in Addis Ababa, most of the patients (42.3%) were visiting ALERT Hospital after 12 months following the appearance of the lesion (Bekele et al. 2014:7). In Turkey majority of the respondents (54.4%) have a lesion with a duration of fewer than six months (Serarslan et al. 2019:113). This may be because of the difference in the health-seeking behaviour of the study community.

Observation was used to determine the location of the lesion, and more than half of the CL cases, 14 (28.57%), acquired a lesion on their cheeks. Multiple lesions were prevalent next to the cheek 9 (18.36%). Likewise, in north Ethiopia majority (39.9%) of the patients were shown to have the lesion on the check (Bisrat et al. 2015:3), and from another report in the same region, lesion on face was observed in 82.1% of the infected populations (Yohannes et al. 2019:6). According to Negera et al. (2008:886), more over half of the CL cases (55.4%) had lesions on one or both cheeks, with 13.0% (12/92) developing lesions on the nose.

Histamine is a pharmacologically active component released from sandflies while feeding to facilitate the feeding process (Kamhawi 2000). Unless disturbed by human activities, sandflies feed at dusk (Adam et al. 2017:15). Sandfly hosts include humans, animals, dogs, rodents, reptiles, amphibians, and birds. The feeding patterns of the same species of sandflies collected from different biotopes are frequently varied, demonstrating that they are opportunistic feeders on easily accessible hosts (Abbasi, Cunio & Warburg 2009). As a result, humans and domestic dogs are likely to be the primary prey for sandflies in urban and peri-urban settings, as feeding is related with increased host census and lower sandfly travel costs to the host (European Centre for Disease Prevention and Control 2020). Humans are preferred hosts for sandfly bites, and bites occur indoors rather than outdoors and in caves, according to a research conducted to understand feeding behavior and activity of *Phlebotomus pedifer* and potential reservoir hosts of *Leishmania aethiopica* in southern Ethiopia (Pareyn et al. 2020:9). This means that humans are becoming more preferred by sandflies and using personal protective

measures (e.g., covering face and hand) while sleeping may prevent the bite from sandflies which are actively searching for a blood meal at night.

In the current study, face parts (nose, ear, lips, cheeks, forehead, chin) were affected by 89.79% (44/49) of the cases. Face parts were also the major location of the lesion in other studies in Ethiopia (Negera et al. 2008:886; Bisrat et al. 2015:3). This may raise a concern of stigmatisation owing to the disfigurement of the face. Carbon dioxide is the single most powerful attractant for blood questing sandflies. This has also been proven for Ethiopian sandflies (Kirstein, Faiman, Gebreselassie, et al. 2013), therefore, bite tends to occur mostly on the face because of CO_2 released while breathing.

Despite the fact that CL has a low morbidity and is not fatal, the ugliness and associated social stigmatization may develop fast psychological disorders and limit the social involvement of those affected by the condition. As a result, CL, like other disfiguring diseases, affects not just the individual's physical well-being but also their psychological, social, and economic well-being (Kassi, Kassi, Afghan, Rehman & Kasi 2008:1). Though little epidemiological research on CL have been conducted in Ethiopia, the moral, attitudinal, psychological, and societal instability that demands significant attention has gone unmentioned. According to a study conducted in Ethiopia to evaluate the psychosocial impact of CL on those infected with the disease, CL can cause a wide range of issues. Including psychological issues (such as low self-esteem, stress, depression, aggression, irritability, frustration, anger, anxiety, hopelessness, dissatisfaction, hostility, guilt, and confusion) as well as social issues (stigma, labeling, discrimination, limited participation, lower quality of life, negative attitudes, poor social relationships, cultural influence, avoidance of marriage ties, rehabilitation center inadequacy and patient hiding) (Semeneh 2012:17). According to the findings of this study, the majority of patients in Turkey (46.76%) have facial lesions (Serarslan et al. 2019:113).

Different lesion nature types were observed in the present study, such as ulcer 20 (40.81%) and nodules with or without crust 22 (44.89%). Most of the lesions were dry 36 (73.46%) and the remaining 13 (26.53%) were wet. Of the wet lesions, seven (14.28%) of the lesions were exudative and draining and others were painful with pruritis. In Addis Ababa, among the different lesions of leishmaniasis, plaque was the most common lesion presented by 50.85% of the cases, followed by an ulcer with nodules, 20.94% (Bekele et al. 2014:7). Lesions were mostly nodular (42.4%) and nodular-ulcerative (23.7%) in Turkey (Serarslan et al. 2019:113).

5.4.4. Smear test result

A total of 49 people with active lesions who agreed to take samples had their skin scrapings were analysed, and amastigotes were found in 34.69% (17/49) of them. This was higher compared to 234 (14.2%) cases diagnosed as positive for CL by direct microscopic examination for amastigote form of Leishmania spp from 1651 patient records suspected and screened for leishmaniasis in Addis Ababa (Bekele et al. 2014:4). Skin scrapings from 43 people with active lesions were carefully analysed in a community-based survey in North Ethiopia, and amastigotes were visible in 69.8% of them (Bisrat et al. 2015:4).

Although smear test with Giemsa staining is cheaper, more accessible, and highly specific, its sensitivity is low for all tissues (53-86%) (Rasti et al. 2016:611). As a result, it was recommended that all lesions lasting less than four months undergo smear testing. The shorter the time since the commencement of the disease, the higher the possibility of a positive result (81.1%), and vice versa (2.7%). (Tareen, Afaq, & Haque 2014:85). However, the majority of patients in this study and another from Addis Ababa had had their lesions for longer than seven months and 12 months, respectively. Adding to this, of the 29 of the individuals who get treated in the current study, 72.4% (21/29) used the application of herbal medicine on the wound which may hamper diagnostic tests.

Six (35.29%) of the 17 smear-positive cases received herbal treatment. The elimination of parasites was not achieved through previously applied herbal

treatment. Despite the fact that several evidence-based in vitro, in vivo, and clinical trial studies have demonstrated the important positive effects of herbal remedies in the treatment of CL lesions (Parvizi, Zare, Handjani, Nimrouzi, & Zarshenas 2020:4), a study conducted in Ethiopia on the efficacy of herbal remedy (Shiunko ointment) suggests that the ointment could be useful as an adjuvant or as a complementary treatment (Na-Bangchang, Ahmed, et al. 2016:1). Furthermore, according to a review of plants used to cure leishmaniasis in Ethiopia, none of the medicinal plants historically used to treat leishmaniasis in Ethiopia have been scientifically validated (Aschale et al. 2018:1).

Bivariate logistic regression test of receiving herbal treatment and smear test revealed that those receiving herbal medication previously are associated with negative results in both smear test [COR= 0.21; 95% CI, 0.057-0.726] and culturing [COR= 0.27; 95% CI, 0.050-1.182]. Negrea et al. (2008:886) pointed out that out of the six individuals with scar, four of them had been treated at the referral dermatology (ALERT Hospital) and the remaining two had recently been treated traditionally at Buie – the administrative town of Sodo. This coupled with smear and culture negativity of samples of individuals previously treated with herbal medicines in this study may indicate parasite reduction ability of the herbal treatments received previously. But since the evidence from this study cannot be used to draw this conclusion, the only assumption made here is that application of herbal medicine has a negative impact on test results. Smears and cultures obtained from patients showed very low or negative results due to fungal contamination of the samples, bacterial superinfection of ulcers, application of local herbs on the lesion, or burning of the lesion, all of which could result in low parasitaemia, according to Bugassa et al. (2014:115).

5.4.5. Culture results

From 49 cultured samples, 18.37% (9/49) were culture positive and 81.63% (40/49) were culture negative in NNN medium. In a community-based survey in North Ethiopia, 10 (32.3%) of the 31 samples cultured were positive (Bisrat et al 2015:4). In a separate study from South Ethiopia, laboratory findings revealed that 4 (13.8

percent) of the 29 patients with active lesions tested positive for the NNN medium (culture) (Bugassa et al. 2014:114). In Brazil, positive cultures in NNN were found in 53 individuals (69.7% of the research participants) (Luz, Silva, Silva, et al. 2009:64). Test techniques should have high sensitivity and specificity, which refers to the ability of the tests to correctly detect false positives and false negatives (Strassle, Hess, Thom, & Harris 2012).

In a retrospective review of the contribution of Leishmania cultures on NNN medium in the Pasteur Institute of Tunisia between 1995 and 2007, the global sensitivity of globally employed NNN culture in the course of CL was determined to be 68.2% (Chouihi, Amri, Bouslimi, et al. 2008). Other studies found NNN medium being less sensitive. Culture using NNN medium was found to be least sensitive as only eight of 55 (14.5%) of the NNN cultures were positive in a comparative study of different culture methods available for leishmaniasis (Hendricks & Wright 1979). Recently conducted similar comparative study also shows that promastigote proliferation was observed in 30.76% of the samples cultured in NNN compared to 92% in other modified culture medium (Liver Extract Medium) (Özbilgin, Tünger, İnanır, et al. 2020:137). In addition to the fact that NNN media has low sensitivity compared to other modified culture media as reported from studies in different countries, cultures obtained from the patients in this study were low because of the use of herbal medicines by the majority of the patients. Similar situations were observed in a study from southern Ethiopia (Bugassa et al. 2014:115).

Out of the nine culture-positive samples in the current study, two (22.2%) were contaminated so deemed unfit for polymerase chain reaction (PCR) analysis. It is known fact that clutures are exposed to contamination specialy ones used for leishmanaisis (Chouihi, et al. 2008). Contamination by bacteria or fungus is a common concern when protozoan parasites like Leishmania are isolated and propagated in vitro (Nuako 2016:41). Antibiotics were added to the NNN media to provide anti-contaminant efficacy while minimizing protozoa toxicity. Contamination rates of 21.1%, on the other hand (Saki, Akhlaghi, Maraghi, et al. 2009:60) and

5.9% (Chouihi, et al. 2008) of NNN media were observed in studies from Iran and Tunisia respectively.

Looking into the clinical forms of nine culture-positive samples, two were from patients who have MCL, and the rest (seven) were from patients with LCL. Of the nine samples positive for culturing on NNN media of skin scrapings, three (33.3%) have received herbal medication this shows that herbal remedies may remove parasites from the site but do not completely heal the person. Several researchers are dedicated to the quest for novel compounds that are effective against Leishmaniasis, according to an integrative study from 2000 to 2011. However, it was clear that the bulk of tests were conducted with the promastigote form under the settings of this investigation (Brito, Santos, Rodrigues, Brito & Xavier-Filho 2013). In a review of Ethiopian medicinal plants, 28 plants were found from diverse research, and CL accounts for a large percentage of Leishmania infection treated with traditional medicinal herbs. However, none of the Ethiopian medicinal plants traditionally used to cure leishmaniasis have been scientifically validated (Aschale et al. 2018:1).

5.4.6. PCR amplification and restriction fragment length polymorphism (RFLP)

All nine culture samples evaluated with PCR for the presence of Leishmania amastigote DNA were found to be positive. The findings are consistent with prior publications (Bsrat et al. 2015:5). When the ITS-1 PCR result was digested with the enzyme Hha I, the patient samples and the L. aethiopica reference strain produced superimposed products of 164 and 162 bp, showing that all of the CL cases were caused by L. aethiopica. In Ethiopia, a similar finding was obtained (Bsrat et al 2015:5; Gadisa, Genetu, Kuru, et al. 2007:341).

5.5. CONCLUSION

The current study's findings were thoroughly described in this chapter. In comparison to other research findings, the community's knowledge, attitude, and practice, variables determining CL transmission, prevalence, and Leishmania

species circulating in the community were discussed. The conclusion and recommendation of the research findings will be presented in the following chapter.

CHAPTER 6

CONCLUSION, LIMITATIONS AND RECOMMENDATIONS OF THE STUDY

6.1. INTRODUCTION

The results of various litratures are discussed in Chapter 5 as well as the study's primary findings. This chapter covers the study's main findings and delivers the research's conclusions in relation to the stated questions and problem statement. In this chapter, the research findings, recommendations, and study contributions are summarised. The study's weaknesses and potential research areas are also discussed.

The purpose of the study was to produce evidence for designing feasible control and prevention strategy of leishmaniasis by gathering epidemiological information on the transmission of CL in Sodo District using a multidisciplinary approach (behavioural, risk factor assessment, and parasite identification). Cognizant of this, the study was conducted in Sodo District, South Ethiopia, and achieved the following objectives:

- To access knowledge, attitude and practices of Sodo community on CL.
- Investigate demographic, household and host-related risks related to transmission of CL.
- To clarify the role of animal and environmental factors on spread of CL in the district.
- To determine the prevalence of CL in the area.
- To identify the Leishmania species circulating in the community.

And generally, to understand key features leishmaniasis transmission in the community and highlight ways forward for the managing CL in Sodo District based on results-driven from objectives above.

6.2. RESEARCH DESIGN AND METHOD

A primary quantitative cross-sectional research design was used in this study. There were three stages to it. The first phase involved a knowledge, attitude, and practice study, followed by a risk factor analysis and a third phase that focused on prevalence estimation and species identification.

The knowledge, attitude, and practice data was collected from 423 household heads in six kebeles using a pre-tested interviewer-administered questionnaire. The process was followed for data quality assurance. Using Access-based Epi-Info 2005 and SPSS version 24.0, data was entered, cleaned, and prepared for analysis. To answer all particular objectives, descriptive, chi-square test, bivariate, and multivariate analyses were used.

The second phase was the analysis of risk factor assessment for the transmission of CL in the community. Data were collected from 1356 individuals using a structured questionnaire. Furthermore, data were subject to bivariate logistic regression and multivariate logistic regression using a backward stepwise approach for selecting most predicting variables in this study and statistical significance was at $p < 0.05$. Multiple correspondence analysis and hierarchal clustering was computed to see the overall interaction between different factors. The statistical analysis was carried out using R Package Version 3.02.

The third phase of the study sought to estimate CL prevalence and isolate the parasite species circulating among the community. Different laboratory methods were used.

In all data collection actions, significant validity and reliability measures were engaged to confirm data quality and reliability. The study firmly followed ethical considerations to defend participants' rights by standard ethical principles.

6.3. SUMMARY AND INTERPRETATION OF THE RESEARCH FINDINGS

This section presents the interpretation and compared findings of the study based on the specific objectives articulated in the results section of this study.

6.3.1. Summary and interpretation of knowledge, attitude and practice survey

A total of 423 individuals were involved in this study; 219 (51.8%) of the respondents were males and 204 (48.2%) were females. Concerning knowledge of CL, 350 (82.7%) of the participants have heard about CL. However, there is a lack of knowledge about the cause of the disease and biting behaviour, breeding site, and preferred biting time of the sandfly vector. This knowledge gap will hinder the adoption of preventive measures and early treatment seeking. Looking into the overall scores of knowledge in the current study, 262 (61.9%) of the respondents have a satisfactory general knowledge of CL. Still, this calls for improvement because important awareness for the prevention of the disease (such as the cause of disease and vector significance) are not well known. Level of education (primary education) and a person’s knowledge of CL previously were significantly associated with satisfactory knowledge.

Regarding attitude of the community towards the disease, 261 (61.7%) of the participants in this study believed that CL can be treated. Communities with unfavourable attitudes of treatment can ignore taking preventative action or visiting health facilities. However, more than half 251 (59.3%) of the respondents preferred indigenous medication through the application of herbs by traditional healers. This is another point of concern since none of the traditionally used medicines in Ethiopia are scientifically proven. Nevertheless, the use of traditional remedies is a necessity rather than a choice since biomedical treatments for leishmaniasis are not available

in the study area. Generally, 226 (53.2%) of the participants have a favourable attitude and this was significantly associated with age, education, and a person's knowledge of CL.

The use of preventive measures was seen among about half (55.6%) of participants. Limited use of personal protective measures, repellents and insecticides exists. On the other contrary, a wider practice of application of herbal remedies was observed. Two hundred thirteen (50.4%) of the respondents have good prevention practice while 210 (49.6%) of the respondents have poor practice. Those who know someone infected with the disease were associated with good practice.

6.3.2. Risk assessment

Among demographic, household, environmental, host, and animal factors, twelve were found to be significantly associated with disease presence. These variables were dumping animal dung near the house, the existence of hyrax, sensel tree and cave gorge near home, presence of screen on window or door of the house, residential kebele (Kola Nurena and Beki). In addition, the variables included the use of personal protective measures, irrigation at night, sleeping covering hand and face, spending time near gorge/cave, sleeping at home yard/in compound and travel history. Another finding is that these factors define or classify the study participants in two distinct groups sharing different characteristics based on disease status on multiple correspondence and hierarchical clustering analysis.

6.3.3. Estimation of disease prevalence and parasite species identification

The overall prevalence of CL in Sodo was 9.07% (123/1356). Of these, 49 (3.61%) were with active CL infection during the visit, and 74 (5.45%) were infected in the past. Active cases of CL were prevalent among individuals who are female, less educated, farmers, in the age group of 5-10 years, and who are from Kola Nurena kebele. Most of the cases are presented with localised CL form. Patients who previously used herbal medication were found positive in the smear test and culture test. Lesions were found primarily on face parts. The isolates identified in this study

are *L. aethiopica*, with a similar RFLP pattern as the *L. aethiopica* reference strain. This result provides evidence for *L. aethiopica* to be the principal aetiologic agent of CL in Sodo.

6.4. CONCLUSION

As a result of the KAP, risk assessment, prevalence estimation, and species identification phases of this investigation, the following findings are drawn. Each study objective's conclusions are presented along with their related research questions.

6.4.1. What is the knowledge attitude and practice of the community?

A satisfactory level of general knowledge was seen among 262 (61.9%) of the respondents. This result may not be considered enough for the prevention of the disease because one-third of the community has unsatisfactory knowledge. Considering the endemicity of the disease in the area, this level of knowledge is deficient especially when there is a knowledge gap on the cause and the vector of the disease which could be the most important knowledge to prevent the disease. Even though the community can recognise CL, transmission methods are poorly understood. This knowledge gap may affect the disease incidence and pose a risk for the endemicity.

The percentage of study participants showing favourable and unfavourable attitudes about CL is 53.4% (n=226) and 46.6 % (n=197) respectively. This compared to other findings is relatively higher. However, there is a concern on attitude about treatment choice of the community as the preferred choice of medication was the application of traditionally known herbs for 251 (59.3%) of the population.

About half 213 (50.4%) of the participants exhibit good preventive practice against the disease while the rest 210 (49.6%) have a poor practice of CL. There is also a bridge in the preventive practice of the community because personal protective measures, use of repellents and application of insecticides are low in addition to the predominant use of herbal remedies for treating the disease.

From the KAP survey results, evidence of discrepancies between knowledge attitude and practice is observed. For example, majority of the respondents (82.7%) have heard about the disease before. About half (51.1%) of them were able to correctly name the disease when shown a picture with a typical case. However, more than half do not know the cause (53.4%) of the disease, 59.4% do not know how the disease is transmitted and 50.4% have no idea about the importance of the vector in disease transmission. Even though 82.7% heard about the disease, more than half (58.6%) think that they are not well informed about CL. Concerning the attitude of respondents about the disease, majority (77.5%) are willing to participate in CL prevention/control programmes and 64.5% think that the disease is very serious in their locality. Nonetheless, only 55.6% have a practice of using preventive measures against the disease.

Comparing overall practice, with knowledge and attitude, there is a statistically significant association between knowledge and practice and again between attitude and practice. Accordingly, the odd of having a good practice is five times higher for those who have satisfactory knowledge than those who have unsatisfactory knowledge, and individuals with favourable attitude are three times more likely to have a good practice.

6.4.2. Which factors play a role in the transmission of leishmaniasis in the area?

Dumping animal dung near the house, the existence of hyrax, sensel tree and cave gorge near home, and the presence of screen on window or door of the house, residential kebele (Kola Nurena and Beki) are factors fueeling the trasmision and the spread of CL. Furthemore, the use of personal protective measures, irrigation at night, sleeping covering hand and face, spending time near gorge/cave, sleeping at home yard/in compound and travel history are factors that play a role in the transmission of CL in the area. Seven out of these factors are host-related factors that are interesting because interventions altering these host behaviours can play a

role in preventing the disease. This research also provides a result that these factors group individuals according to their disease status. Therefore, interventional investments targeting these factors can ultimately affect disease outcome and prevention in the community.

6.4.3. What is the burden of the disease in Sodo District?

The overall prevalence of CL in the area was 9.07% (123/1,356) for both scar and active infection. Of these, 49 (3.61%) were with active CL infection during the visit, and 74 (5.45%) were infected in the past (with scar). This result is comparable with other studies in the country and is of considerable importance since the reported prevalence of active CL in the country ranges between 1.53% to 6.70%. There is a difference in the proportion of active CL between different residential kebele, gender, age, and educational status. The most common form of CL among cases was LCL and lesions are mainly located on face parts. Even though those who received herbal medication are inversely associated with smear and culture test positivity, some of these patients were also found positive in the smear test and culture test. This means that the herbal medication applied may reduce the parasite but does not completely remove it from the body.

6.4.4. What species of Leishmania is circulating in the community?

This result provides evidence for *L. aethiopica* to be the principal aetiologic agent of CL in Sodo which is the first report in the area to the researcher's knowledge.

6.5. RECOMMENDATIONS

This section contains recommendations based on the study's findings at various levels that are thought to be relevant to use and implement the recommendations.

6.5.1. Recommendations from KAP survey

Recommedations drived from the study findings are:

- Awareness of the community needs to be improved especially on cause and transmission means of the disease.

- Comprehensive effort must be exerted to increase community awareness on the importance of sandfly vector including breeding places, resting sites, and preferred biting time so that the community will invest in disease control by denying access to such breeding and resting sites within the household and the surrounding environments.

- The perception of the community also need to be transformed because they have a low habit of utilisation of modern medication. Yet, the preference for herbal medicine is not a choice but a necessity owing to the unavailability of CL treatment in the study area. Therefore, there is a need to provide CL treatment readily available to the community and to investigate locally applied herbs for their efficacy. Since there is no national CL treatment protocol in the country and available drugs have complications and need a long duration of treatment, the use of locally available medicinal plants will be beneficial because if proven effective, the use of herbal remedies is already a widely accepted and practiced approach by the community.

- The community needs to be sensitised about the use of preventive measures especially personal protective measures that could create a barrier to sandfly bite, use of repellents and insecticide application. Because prevention is preferable to cure, community support for the implementation of protective measures against CL is critical. With the motto "Small bite, big threat," WHO (2014) emphasised the serious and growing threat of vector-borne illnesses around the world, particularly leishmaniasis. In addition, WHO (2014) established a network for CL control with the objective of decreasing disease transmission by addressing all aspects involved in the cycle (e.g., vector and reservoir control; preventive measures).

- Community outreach on CL will have a huge advantage as it has been shown from the findings of these research those who know someone infected with CL proven to have better knowledge, attitude, and practice. In addition, it has also been noticed that satisfactory knowledge and favourable attitude have an association with good practice. Over and above, the importance of awareness education in disease prevention has been shown

to be effective in many studies.

- The above-recommended awareness creation programmes should specifically target individuals of different ages and levels of education.

6.5.2. Recommendations from the risk assessment survey

- The habit of dumping animal dung (cow) should be kept far from the house. Ethiopians in rural areas use cow dung for house beautification and cleaning. They also make a cake from cow dung called “kubet” in Amharic being dried to be used as fuel for cooking food by women. Because adult sandflies are weak fliers with a characteristic short hopping flight, they can only move a few hundred meters away from their nesting places, the kubet should be kept at least half a kilometer away from the home (Braks, Giuseppe & Goffredo 2017:20).

- Favourable conditions to exitance of reservoir host hyraxes (such as cave or gorge) or places that serve as resting sites for sandfly vector (sensel tree) around households should be monitored. Accompanying these monitoring actions, the community needs to be aware of the danger of being near these places.

- Specific CL control and prevention programme needs to be adopted locally because it is difficult to integrate the CL control programme with a malaria control programme owing to different geographical distribution (CL is present in malaria-free highland areas). For example, Kola Nurena and Beki kebeles are considered non-endemic to malaria but these kebeles harboured majority of CL cases.

- When designing CL control and prevention programmes, priority should be given to host behavior related factors such as putting a screen on window or door of the house, use of personal protective measures, irrigation at night, sleeping covering hand and face, spending time near gorge/cave and sleeping at home yard/in the compound. Investing in alteration of human

behaviours would be cost-effective for many reasons. For once, out of significant factors affecting disease distribution, majority attribute to human behaviour. Again, modifying environmental factors (cave, gorge, trees) and animal factors (hyrax) require a big budget, is not environmentally friendly, and may not be sustainable. On some occasions, driving hyraxes away from human proximity may make humans more exposed to sandfly bite as research shows that sand flies are opportunistic feeders that may target humans nearby when hyraxes are not available. A study conducted to understand the feeding behaviour of Ethiopian sandflies also indicated that sandflies do not make a preference between humans and hyrax (Pareyn et al. 2020:9).

- From the risk assessment study, priority areas for investments are on awareness creation to avoid dumping animal dung near home. In addition, people need to stop irrigating crops at night, use personal protective measures, sleep hand/face covered, stop sleeping at the home yard, and put screen on window or door coupled with environmental management. The latter include insecticide spraying of cave/gorges, sensel trees areas, and other hyrax borrows and sandfly resting places near residential areas to prevent the community from getting infected.

6.5.3. Recommendations from prevalence study and species identification study

- CL is a challenging public health problem in the area indicated by the prevalence rates of active cases and scars. Therefore, due attention needs to be given to it.

- The community needs to be educated about using personal protective measures like covering face and hand while sleeping and wearing appropriate clothing (long sleeves) as lesions are common on face parts.

- A thorough investigation of the efficacy of herbal remedies commonly used among the community needs to be done because the disease was confirmed in those who received the treatment.

- The Leishmania species circulating in the community is *L. aethiopica* which has particularly diverse manifestations and is pleotropic as well. *Since L. aethiopica* is mostly found in Ethiopia and, to a lesser extent, Kenya. As a result, treatment plans developed for other species that have been well examined may not be useful in treating *L. aethiopica*. Furthermore, the frequent occurrence of DCL and MCL in L. aethiopica suggests that systemic therapy may be required more frequently than in other species such as L. major or L. tropica. As a result, randomised clinical trials are desperately needed and should be carried out to help find possibly effective CL medicines.

6.7. CONTRIBUTIONS OF THE STUDY

- Contribution of knowledge, attitude, and practice survey: Gaps in knowledge attitude and practice and risk factors associated with these can help inform awareness creation programmes to help prevent and control the disease in the area. Awareness creation strategies can hasten modification of behavioural patterns of the Sodo communities.

- Contribution of risk factor assessment survey: The demographic, household, environmental, host-related, and animal-related factors identified as risk factors of CL in this study could help implementation of control strategies. Besides, findings also provide information on the effectiveness of targeting these factors as they can determine disease outcomes. In limited-resource settings such as Sodo identification of implementation priorities is of vital importance.

- Contribution of prevalence study and species identification study: Magnitude of the disease, population at risk and species of the parasite in

the study area were not known before. This information will be the foundation for launching successful CL management in the area, in addition to providing as a baseline for future investigations. The size of CL in the study area is given, which will aid zonal and woreda health bureaus in planning efforts and human resources to reduce disease burden. Some of the disease's primary potential determinants have been identified, which will aid in the formulation and execution of control programs. The most common Leishmania species in the area have been identified, which will aid in breaking the transmission cycle and determining the best treatment choices.

6.8. LIMITATIONS OF THE STUDY

With the research design being cross-sectional, the study is subject to disadvantages associated with this design. Different from other studies, cross-sectional studies require larger sample sizes since the whole population is being studied at once (Pourhoseingholi, Vahedi & Rahimzadeh 2013). When a small sample is used, the risk of error rises considerably since the results could be due only to chance or coincidence. However, in this study, the KAP and risk factor assessment/prevalence studies each included 16.0% of the total household and 8.7% of the overall population, respectively. Cross-sectional studies are intended to give correlated data that can be utilised to draw conclusions about specific populations. Cross-sectional studies are limited to identify causal relationships (Gaille 2018). Therefore, no causal relationship is established in the current study.

The study has been conducted only in Sodo District and findings may not represent other districts nearby or the southern part of Ethiopia at large. Another limitation of this study is the absence of epidemiological studies on the reservoir hosts and vectors which also could provide valuable information to capture the full picture of transmission dynamics of CL in the area.

6.9. FUTURE RESEARCH

- Discrepancies in knowledge, attitude, and practice survey require further examination.

- Because the snapshot character of cross-sectional research, while handy, does not provide a good basis for demonstrating causation, the risk factor assessment study should be based on longitudinal studies or random controlled trials.

- The role of reservoir hosts and detailed investigation of sandfly vectors needs investigation.

- Detection and evaluation of the efficacy of herbal medicines used locally with pinpointing ways of improving CL treatment services with herbal medicines need research.

6.10. CONCLUDING REMARKS

Regardless of limitations, this study provided concrete information on gaps in the knowledge attitude and practice of Sodo community regarding CL. Major factors playing a role in disease transmission, disease magnitude, characteristics of exposed individuals, and Leishmania species were presented. This research, being the first of its kind in the area, provides evidence that can help policymakers, health professionals, intervention programs, researchers, and other stakeholders.

REFERENCES

Abazid, N, Jones, C & Davies, CR. 2012. Knowledge, attitudes, and practices about leishmaniasis among cutaneous leishmaniasis patients in Aleppo, Syrian Arab Republic. *Eastern Mediterranean Health Journal*. 18(1): 7–14. doi.org/10.26719/2012.18.1.7.

Abbasi, I, Cunio, R, & Warburg, A. 2009. Identification of blood meals imbibed by phlebotomine sand flies using cytochrome b PCR and reverse line blotting. *Vector-Borne and Zoonotic Diseases*. 9(1):79–86. doi.org/10.1089/vbz.2008.0064.

Aberra, L, Abera, A, Belay, T, Kebede, A, Gadisa, E & Tasew, G. 2019. Evaluation of microcapillary culture method for the isolation of Leishmania aethiopica parasites from patients with cutaneous lesions in Ethiopia. *Diagnostic and Prognostic Research*. 3(1):1–8. doi.org/10.1186/s41512-019-0051-z.

Adam, AA, Hassan, BM, M, Abdelnour, MO, Awadallah, HA. 2017. Identification and Classification of Sand Flies Species and It's Habitats in El-Kadaba Village, White Nile State, Sudan. *International Journal of Infectious Diseases and Therapy*. 2(1):15–21. doi.org/10.11648/j.ijidt.20170201.14.

Akhoundi, M, Kuhls, K, Cannet, A, Votýpka, J, Marty, P, Delaunay, P & Sereno, D. 2016. A Historical Overview of the Classification, Evolution, and Dispersion of Leishmania Parasites and Sandflies. *PLoS Neglected Tropical Diseases*. 10(3):1–40. doi.org/10.1371/journal.pntd.0004349.

Aklilu, E, Gebresilassie, A, Yared, S. *et al.* 2017. Comparative study on the nocturnal activity of phlebotomine sand flies in a highland and lowland foci of visceral leishmaniasis in north-western Ethiopia with special reference to *Phlebotomus orientalis*. *Parasites Vectors* 10(1):393. https://doi.org/10.1186/s13071-017-2339-6

Akram, A, Khan, HAA, Qadir, A & Sabir, AM. 2015. A cross-sectional survey of knowledge, attitude and practices related to cutaneous leishmaniasis and sand flies in Punjab, Pakistan. *PLoS ONE*. 10(6):1–8. doi.org/10.1371/journal.pone.0130929.

Alemayehu, B & Alemayehu, M. 2017. Leishmaniasis: A Review on Parasite, Vector and Reservoir Host. *Health Science Journal*. 11(4):1–6. doi.org/10.21767/1791-809x.1000519.

Alemu, A, Alemu, A, Esmael, N, Dessie, Y, Hamdu, K, Mathewos, B & Birhan, W. 2013. Knowledge, attitude, and practices related to visceral leishmaniasis among residents in Addis Zemen town, South Gondar, Northwest Ethiopia. *BMC Public Health*. 13(1):382. doi.org/10.1186/1471-2458-13-382.

Alhaj, S. 2018. Available from: https://www.linkedin.com/pulse/kap-survey-how-develop-knowledge-attitude-practice-alhaj-/. (Accessed on 7 July 2020).

Alidadi S, Oryan A. 2014. Cutaneous Leishmaniasis and the Strategies for Its Prevention and Control. *Tropical Medicine & Surgery*. 02(02):35-40. doi.org/10.4172/2329-9088.1000e114.

Alkulaibi, MM, Suleiman, AM, Gasim, EA & Al-Garadi, MA. 2019. Prevalence of Cutaneous Leishmaniasis in Western Highlands in Yemen. *Journal of Tropical Medicine*. 2019: 8248916. doi.org/10.1155/2019/8248916.

Alvar, J, Yactayo, S & Bern, C. 2006. Leishmaniasis and poverty *Trends in parasitology*, 22(12):552-557. doi.org/10.1016/j.pt.2006.09.004.

Alves, F, Bilbe, G, Blesson, S, Goyal, V, Monnerat, S, Mowbray, C, Muthoni, G. et al. 2018. Recent Development of Visceral Leishmaniasis Treatments: Successes, Pitfalls, and Perspectives. *Clinical Microbiology Reviews*. 31(4):1–30. doi.org/10.1128/CMR.00048-18

Amin, TT, Kaliyadan, F, Al-Ajyan, MI, Al-Arfaj, AK, Al-mujhim, MA, Al-Harbi, SJ, *et al.* 2012. Public awareness and attitudes towards cutaneous leishmaniasis in an endemic region in Saudi Arabia. *J Eur Acad Dermatol Venereol*, 26:1544-1551. doi.org/10.1111/j.1468-3083.2011.04339.x.

Anderson L A. 2021. Insect Repellents: Safe and Effective Use. www.drugs.com/article/how-to-safely-use-insect-repellents.html. (Accessed on January 2, 2021).

Arana, BA, Rizzo, NR, Navin, TR, Klein, RE & Kroeger, A. 2000. Cutaneous leishmaniasis in Guatemala: people's knowledge, concepts, and

practices. *Annals of Tropical Medicine & Parasitology*, *94*(8):779-786. doi.org/10.1080/00034900200124l6.

Aryal S. 2019. Source and Reservoir of Infection. Microbe Notes. Online Microbiology and Biology Study Notes https://microbenotes.com/source-and-reservoir-of-infection/ (Assessed on June 19,2019).

Aschale, Y, Wubetu, M & Reta, H. 2018. Ethnobotany of Medicinal Plants Used to Treat Leishmaniasis in Ethiopia: A Systematic Review. *Journal of Traditional Medicine & Clinical Naturopathy*. 07(02):2–7. doi.org/10.4172/2573-4555.1000271.

Assefa, A. 2018. Leishmaniasis in Ethiopia: A systematic review and meta-analysis of prevalence in animals and humans. *Heliyon*. 4(8): e00723. doi.org/10.1016/j.heliyon. 2018.e00723.

Awosan, K, Isah, B, Alayande, M, Enokela, E, Makusidi, M, Agwu, Ngwobia, P & Abubakar, U, Abdullahi, Z & Aderahman, A. 2013. Knowledge, attitude and practice related to management of cutaneous leishmaniasis among physicians in tertiary healthcare facilities in Sokoto, Nigeria. *Global Advanced Research Journal of Medicine and Medical Sciences (GARJMMS).* 2(12):256-263. (ISSN: 2315-5159).

Babbie, ER. 2010. The Practice of Social Research. 12th ed. Belmont, CA: Wadsworth Cengage. https://b-ok.cc/book/1220723/70f1f0. (Accessed on 21 May 2019).

Banerjee, A, & Chaudhury, S. 2010. Statistics without tears: Populations and samples. *Industrial Psychiatry Journal*, *19*(1):60–65. doi.org/10.4103/0972-6748.77642.

Bari, AU. 2012. Clinical spectrum of cutaneous leishmaniasis: An overview from Pakistan. *Dermatology Online Journal*, 18(2):4. PMID: 22398225.

Bekele, S, Bekele, Y, Mulatu, F, Lemma, T, Tilahun, H, Gadisa, E, Negussie, S, Yamuah, L, et al. 2014. Recent trends of cutaneous leishmaniasis in alert hospital, Addis Ababa. *Ethiopian Medical Journal*. (SUPPL. 1):37–41. PMID: 24696987.

Belo, VS, Werneck, GL, Barbosa, DS, Simões, TC, Nascimento, BWL, da Silva, ES & Struchiner, CJ. 2013. Factors Associated with Visceral Leishmaniasis

in the Americas: A Systematic Review and Meta-Analysis. *PLoS Neglected Tropical Diseases*. 7(4): e2182. doi.org/10.1371/journal.pntd.0002182.

Berhe, M, Bsrat, A, Taddele, H, Gadissa, E, Hagos, Y, Tekle, Y & Abera, A. 2018. Knowledge Attitude and Practice towards Visceral Leishmaniasis among Residents and Health Professionals in Welkait District, Western Tigray, Ethiopia. *Journal of Tropical Diseases,* 06(01):1-8. doi.org/10.4172/2329-891x.1000257.

Berkessa, T, Oljira, D & Tesfa, B. 2016. Insecticide treated nets use and its determinants among settlers of Southwest Ethiopia Global health. *BMC Public Health*. 16(1):1–8. doi.org/10.1186/s12889-016-2768-8.

Bern, C, Courtenay, O & Alvar, J. 2010. Of cattle, sand flies, and men: A systematic review of risk factor analyses for South Asian visceral leishmaniasis and implications for elimination. *PLoS Neglected Tropical Diseases*. 4(2): e599 doi.org/10.1371/journal.pntd.0000599.

Bern, C, Joshi, AB, Jha, SN, Das, ML, Hightower, A, Thakur, GD & Bista, MB. 2000. Factors associated with visceral leishmaniasis in Nepal: Bed-net use is strongly protective. *American Journal of Tropical Medicine and Hygiene*. 63(3–4):184–188. doi.org/10.4269/ajtmh.2000.63.184.

Bi, K, Chen, Y, Zhao, S, Kuang, Y & John Wu, CH. 2018. Current Visceral Leishmaniasis Research: A Research Review to Inspire Future Study. *BioMed Research International*. 2018 :9872095. doi.org/10.1155/2018/9872095.

Bisetegn, H, Zeleke, AJ, Gadisa, E, Shumie, G, Damte, D, Fenta, T, Behaksra, S & Bayih, AG. 2020. Clinical, parasitological, and molecular profiles of Cutaneous Leishmaniasis and its associated factors among clinically suspected patients attending Borumeda Hospital, North-East Ethiopia. *PLoS Neglected Tropical Diseases*. 14(8): e0008507. doi.org/10.1371/journal.pntd.0008507.

Bock T. 2020. What is a Dendrogram? Displayer Blog. www.displayr.com/what-is-dendrogram/ (Accessed on February 2,2020).

Boru, G, Shimels, CT & Bilal, AI. 2017. Factors contributing to non-adherence with treatment among TB patients in Sodo Woreda, Gurage Zone, Southern Ethiopia: A qualitative study. *Journal of Infection and Public Health*. 10(5):527–533. doi.org/10.1016/j.jiph.2016.11.018.

Braks, M, Giuseppe, M & Goffredo, M. 2017. Risk of vector-borne diseases for the EU: Entomological aspects – Part 1. External Scientific Report. EFSA Supporting Publications. 14:51. doi: 10.2903/sp.efsa.EN-1173

Brito, AM, Dos Santos, D, Rodrigues SA, Brito, RG, & Xavier-Filho, L. 2013. Plants with anti-Leishmania activity: Integrative review from 2000 to 2011. *Pharmacogn Rev*. 2013;7(13):34-41. doi:10.4103/0973-7847.112840

Bsrat, A, Berhe, N, Balkew, M, Yohannes, M, Teklu, T, Gadisa, E, Medhin, G & Abera, A. 2015. Epidemiological study of cutaneous leishmaniasis in Saesie Tsaeda-emba district, eastern Tigray, northern Ethiopia. *Parasites and Vectors*. 8(1):1–9. doi.org/10.1186/s13071-015-0758-9.

Bugssa, G, Hailu, A, Demtsu, B. 2014. The Current Status of Cutaneous Leishmaniasis and the Pattern of Lesions in Ochollo Primary School Students, Ochollo, Southwestern Ethiopia. *Science Journal of Clinical Medicine*. 3 (6):111-116. doi: 10.11648/j.sjcm.20140306.13.

Calderon-Anyosa, R, Galvez-Petzoldt, C, Garcia, PJ & Carcamo, CP. 2018. Housing characteristics and leishmaniasis: A systematic review. *American Journal of Tropical Medicine and Hygiene*. 99(6):1547–1554. doi.org/10.4269/ajtmh.18-0037.

Center for Disease Control. 2020. Parasites – Leishmaniasis. https://www.cdc.gov/parasites/leishmaniasis/biology.html. (Accessed on 18 January 2020).

Central statistical Agency. 2007. Population and Housing Census of Ethiopia: Statistical SNNPR, CSA, 2012. www.csa.gov.et/newcsaweb/images/.. 2007/. /Reports/. /Statistical SNNPR PartI.pdf. (Accessed on Sep 18,2020).

Central Statistical Agency of Ethiopia. 2012. Population Projections for Ethiopia 2007-2037. https://www.statsethiopia.gov.et/population-projection/ (Accessed on 18 January 2020).

Central Statistical Agency of Ethiopia. 2014. *Summary and Statistical Report of the 2007 Population and Housing Census Results.* Addis Ababa, Ethiopia: Population and Housing Census Commission 57–60. www.ethiopianreview.com. (Assessed on Oct 27, 2020).

Centre for Disease control. 2019. Insecticide-Treated Bed Nets. https://www.cdc.gov/malaria/malaria_worldwide/reduction/itn.html. (Assessed on Oct 27, 2020).

Centre for Disease control. 2020. Leishmaniasis biology. https://www.cdc.gov/parasites/leishmaniasis/biology.html.(Assessed on Oct 27, 2020).

Choueiry, G. 2020. Understanding forward and backward stepwise regression. https://quantifyinghealth.com/stepwise-selection/(Accessed on Sep 19,2020).

Chouihil, E, Amri, F, Bouslimi, N, Siala, E, Selmi, K, Zallagua, N, Abdallah, RB, Bouratbine, A, & Aoun, K. 2009. Cultures on NNN medium for the diagnosis of leishmaniasis. *Pathologie Biologie*, 57(3):219-24. DOI: 10.1016/j.patbio.2008.03.007.

Costa, VMS, Marques, SR, Ramos, JV, Santana, IM, Silva, CM, Alves, LC & Faustino, MA. 2013. Health education for control of leishmaniasis in the endemic area. 7. 591-592. https://www.researchgate.net/publication/290601651_Health_education_for_control_of_leishmaniasis_in_endemic_area. (Accessed on February 2,2019).

Coura-Vital, W, Marques, MJ, Veloso, VM, Roatt, BM, Aguiar-Soares, RD, Reis, LE, Braga, S, Morais, MH, Reis, AB, & Carneiro, M. 2011. Prevalence and Factors Associated with Leishmania infantum Infection of Dogs from an Urban Area of Brazil as Identified by Molecular Methods. *PLoS Neglected Tropical Diseases.* 5 (8). doi.org/10.1371/journal.pntd.0001291.

Creswell, JW & Creswell, JD. 2018. *Research Design: Qualitative, Quantitative and Mixed Methods Approaches*. 5th Edition. London: SAGE Publications Inc.

Cruncic, A. 2019. Understanding enternal and external validity. How These Concepts Are Applied in Research Available from: https://www.verywellmind.com/internal-and-external-validity-4584479. (Accessed on 2 November 2020).

Cutler, DM & Lleras-Muney, A. 2006. *Education and Health: Evaluating Theories and Evidence*. doi.org/10.3386/W12352.

Dassoni, F, Daba, F, Naafs, B & Morrone, A. 2017. Leishmaniasis recidivans in Ethiopia: Cutaneous and mucocutaneous features. *Journal of Infection in Developing Countries*. 11(1):106–110. doi.org/10.3855/jidc.8516.

Davis, P 2019. Leishmaniasis facts. https://www.emedicinehealth.com/leishmaniasis/article_em.htm. (Accessed on 29 Oct 2020).

Dawit, G & Shishay, K. 2014. Epidemiology, public health impact and control methods of the most neglected parasite diseases in Ethiopia: A review. *World Journal of Medical Sciences*. 10(2):94–102. doi.org/10.5829/idosi.wjms.2014.10.2.81231.

Dawit, G, Girma, Z & Simenew, K. 2013. A Review on Biology, Epidemiology, and Public Health Significance of Leishmaniasis. *Journal of Bacteriology & Parasitology*. 04(02). doi.org/10.4172/2155-9597.1000166.

Degu, GE. 2006. Cutaneous Leishmaniasis in Saris, Addis Ababa. A thesis submitted to the School of Graduate Studies: In partial fulfillment of the requirements for the Degree of Master of Science in Biology. Obtained from Personal communication.

Deribe, K, Meribo, K, Gebre, T, Hailu, A, Ali, A, Aseffa, A & Davey, G. 2012. The burden of neglected tropical diseases in Ethiopia, and opportunities for integrated control and elimination. *Parasites and Vectors*. 5(1):1–15. doi.org/10.1186/1756-3305-5-240.

Desta, A, Shiferaw, S, Kassa, A, Shimelis, T & Dires, S. 2005. Leishmaniasis For the Ethiopian Health Center Team.

Doe, ED, Egyir-Yawson, A & Kwakye-Nuako, G. 2019. Knowledge, Attitude and Practices Related to Cutaneous Leishmaniasis in Endemic Communities

in the Volta Region of Ghana. *International Journal of Healthcare Sciences*. 7(1):33–43. ISSN 2348-5728.

Dohoo I, Martin W, Stryhn H. 2014. *Veterinary Epidemiologic Research*. Model building strategies. 2nd edition. Canada. VER Inc. page 369. doi.org/10.1155/2016/5984709.

Dvorak, V, Halada, P, Hlavackova, K, Dokianakis, E, Antoniou, M & Volf, P. 2014. Identification of phlebotomine sand flies (Diptera: Psychodidae) by matrix-assisted laser desorption/ionization time of flight mass spectrometry. *Parasites and Vectors*. 7(1):1–7. doi.org/10.1186/1756-3305-7-21.

Elnaiem, DE, Khogali, A, Alsharif, B, Dakein, O, Jibreel, T, Hassan, M, Edries, HH, Elhadi, H, et al. 2020. Understanding sand fly sampling methods: Sticky traps are attraction-based and not interceptive sampling tools of *Phlebotomus* orientalis. *Parasites and Vectors*. 13(1):1–15. doi.org/10.1186/s13071-020-04249-1.

Engels, D & Zhou, XN. 2020. Neglected tropical diseases: An effective global response to local poverty-related disease priorities. *Infectious Diseases of Poverty*. 9(1):1–9. doi.org/10.1186/s40249-020-0630-9.

Erber, AC, Arana, B, Ben, SA, Bennis, I, Boukthir, A, Castro Noriega MdM, et al. 2020. Patients' preferences of cutaneous leishmaniasis treatment outcomes: Findings from an international qualitative study. *PLoS Negl Trop Dis* 14(2): e0007996. https://doi.org/10.1371/journal. pntd.0007996.

Ertabaklar, H, Çalişkan, SÖ, Boduç, E & Ertuğ, S. 2015. Comparison of Direct Microscopy, Culture and Polymerase Chain Reaction Methods for the Diagnosis of Cutaneous.*Mikrobiyol Bul.* 49(1):77-84. doi: 10.5578/mb.8344. PMID: 25706733.

Eshetu, B & Mamo, H. 2020. Cutaneous leishmaniasis in north-central Ethiopia: trend, clinical forms, geographic distribution, and determinants. *Tropical Medicine and Health*. 48(1):39. doi.org/10.1186/s41182-020-00231-w.

Ethiopian Neglected Tropical Disease master plan. 2016. Second Edition of National Neglected Tropical Diseases Master Plan

https://espen.afro.who.int/system/files/content/resources/ETHIOPIA_NTD_Master_Plan_2016_2020.pdf .(Assessed on 7 March 2020).

European Centre for Disease Prevention and Control.2020. Phlebotomine sand flies - Factsheet for experts. https://www.ecdc.europa.eu/en/disease-vectors/facts/phlebotomine-sand-flies.(Assessed on 7 March 2020).

Fateh, K, Vatandoost, H, Rassi,Y, Naseh, M, Mehdi, M, Mohammad, HS, Mona, K, et al. 2019. Aerobic midgut microbiota of sand fly vectors of zoonotic visceral leishmaniasis from northern Iran, a step toward finding potential paratransgenic candidates. *Parasites & Vectors*. 12(1):10. https://doi.org/10.1186/s13071-018-3273-y.

Federal Democratic Republic of Ethiopia Ministry of Health. 2005. National Hygiene and Sanitation Strategy. For To Enable 100 % Adoption of Improved Hygiene and Sanitation.https://www.wsp.org/sites/wsp/files/publications/622200751450_EthiopiaNationalHygieneAndSanitationStrategyAF.pdf. (Accessed on 18 July 2019).

Federal Ministry of Health. 2020. Malaria Prevention & Control Program. Available from: http://www.moh.gov.et/ejcc/en/malaria-prevention-control-program**.** (Assessed on 29 Oct 2020).

Fenethun, Y, Eshetu, G, Worku, A, Abdella, T, Yeneayehu Fenetahun, C & Fenetahun, Y. 2017. A survey on medicinal plants used by traditional healers in Harari regional State, East Ethiopia. *Journal of Medicinal Plants Studies*. 5(1):85–90. ISSN: 2320-3862

Fit for travel .2020. Information on how to stay safe and healthy abroad. Leishmaniasis. https://www.fitfortravel.nhs.uk/advice/disease-prevention-advice/leishmaniasis (Assessed on 29 Oct 2020).

Fitzpatrick, CNU, Lenk, U, de Vlas, SJ, Bundy, DAP.2017. An Investment Case for Ending Neglected Tropical Diseases. In: *Disease Control Priorities* (third edition), vol. 6. Washington (DC): The International Bank for Reconstruction and Development / The World Bank. DOI: 10.1596/978-1-4648-0524-0_ch17.

Fritzell, C, Raude, J, Adde, A, Dusfour, I, Quenel, P & Flamand, C. 2016. Knowledge, Attitude and Practices of Vector-Borne Disease Prevention during the Emergence of a New Arbovirus: Implications for the Control of Chikungunya Virus in French Guiana. *PLoS Neglected Tropical Diseases*. 10(11):1–18. doi.org/10.1371/journal.pntd.0005081.

Gadisa, E, Genetu, A, Kuru, T, Jirata, D, Dagne, K, Aseffa, A & Gedamu, L. 2007. Leishmania (Kinetoplastida): Species typing with isoenzyme and PCR-RFLP from cutaneous leishmaniasis patients in Ethiopia. *Experimental Parasitology*. 115(4):339–343. doi.org/10.1016/j.exppara.2006.09.014.

Gadisa, E, Tsegaw, T, Abera, A, Elnaiem, DE, Den Boer, M, Aseffa, A & Jorge, A. 2015. Eco-epidemiology of visceral leishmaniasis in Ethiopia. *Parasites and Vectors*. 8(1):1–10. doi.org/10.1186/s13071-015-0987-y.

Gaille, B. 2018. 15 Cross Sectional Study Advantages and Disadvantages. Brandongaille. https://brandongaille.com/15-cross-sectional-study-advantages-and-disadvantages/(Accessed on Dec 26, 2020).

Garapati, P, Pal, B, Siddiqui, NA, Bimal, S, Das, P, Murti, K & Pandey, K. 2018. Knowledge, stigma, health seeking behaviour and its determinants among patients with post kalaazar dermal leishmaniasis, Bihar, India. *PLoS ONE*. 13(9):1–13. doi.org/10.1371/journal.pone.0203407.

Gebremichael, D. 2018. Zoonotic impact and epidemiological changes of leishmaniasis in Ethiopia. *Open Veterinary Journal*. 8(4):432–440. doi.org/10.4314/ovj. v8i4.13.

Gedda, MR, Singh, B, Kumar, D, Singh, AK, Madhukar, P, Upadhyay, S, Singh, OP & Sundar, S. 2020. Post kala-azar dermal leishmaniasis: A threat to elimination program. *PLoS Neglected Tropical Diseases*. 14(7):1–25. doi.org/10.1371/journal.pntd.0008221.

Ghatee, MA, Taylor, WR & Karamian, M. 2020. The Geographical Distribution of Cutaneous Leishmaniasis Causative Agents in Iran and Its Neighboring Countries, A Review. *Frontiers in Public Health*. 8:11. doi.org/10.3389/fpubh.2020.00011.

Ghezzi, P. 2020. Environmental risk factors and their footprints in vivo – A proposal for the classification of oxidative stress biomarkers. *Redox Biology*. 34:101442. https://doi.org/10.1016/j.redox.2020.101442.

Giménez-Ayala, A & Britez, Nilsa, Rojas de Arias, A & Ruoti, M. 2018. Knowledge, attitudes, and practices regarding the leishmaniases among inhabitants from a Paraguayan district in the border area between Argentina, Brazil, and Paraguay. *Journal of Public Health*. 26:1-10. Doi.org/10.1007/s10389-018-0908-6.

Glans, H, Dotevall, L, Söbirk, SK. *et al.* 2018. Cutaneous, mucocutaneous and visceral leishmaniasis in Sweden from 1996–2016: a retrospective study of clinical characteristics, treatments, and outcomes. *BMC Infect Dis*. 18:632. https://doi.org/10.1186/s12879-018-3539-1.

Global Burden of Disease. 2015. Mortality and Causes of Death Collaborators.: Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: a systematic analysis for the Global Burden of Disease Study. *Lancet*, 388:1459–1544. doi: 10.1016/S0140-6736(16)31012-1.

Gonzalez AM, Solís-Soto MT, Radon K. 2017. Leishmaniasis: who uses personal protection among military personnel in Colombia? *Annals of Global Health* 83(3–4):519-523. Doi:/10.1016/j.aogh.2017.10.015.

Gottwalt, A. 2013. Impact of deforestation on vector-borne disease incidence. *J. Glob. Health*. 3(2):16–19. https://doi.org/10.7916/thejgh.v3i2.4864.

Gradoni, L, Lopez-Velez, R & Mourad, M. 2017. Manual on case management and surveillance of the leishmaniases in the WHO European Region. WHO Regional Office for Europe. ISBN 978 92 89052 51 1. Available at https://www.who.int/leishmaniasis/resources/978-92-89052-51-1/en/(Accessed on December 12, 2020).

Griensven, J, Gadisa, E, Aseffa, A, Hailu, A, Beshah, AM & Diro, E. 2016. Treatment of Cutaneous Leishmaniasis Caused by Leishmania aethiopica: A Systematic Review. *PLoS Neglected Tropical Diseases*. 10(3): e0004495. doi.org/10.1371/journal.pntd.0004495.

Guideline for diagnosis, treatment & prevention of leishmaniasis in Ethiopia. 2013. https://www.afrikadia.org/wpcontent/uploads/2018/08/VL_Guidelines_Ethiopia_2013.pdf. (Accessed on Dec 26, 2020).

Gyapong, J & Boatin, B. 2016. Neglected Tropical Diseases. ISBN 978-3-319-25469-2. DOI 10.1007/978-3-319-25471-5. Springer Cham Heidelberg New York Dordrecht London. Springer International Publishing Switzerland 2016.

Hamid, JS, Meaney, C, Crowcroft, NS, Granerod, J & Beyene, J. 2010. Cluster analysis for identifying sub-groups and selecting potential discriminatory variables in human encephalitis. *BMC Infectious Diseases*. 10(1):364. doi.org/10.1186/1471-2334-10-364.

Handler, MZ, Patel, PA, Kapila, R, Al-Qubati, Y, Schwartz, RA. 2015. Cutaneous and mucocutaneous leishmaniasis: Clinical perspectives. *J Am Acad Dermatol*. 73(6):897-908. doi: 10.1016/j.jaad.2014.08.051. PMID: 26568335.

Hejazi, SH, Hazavei, SMM, Bidabadi, LS, Shademani, A, Siadat, AH, Zolfaghari-Baghbaderani, A, Nilforoushzadeh, MA & Hosseini, SM. 2010. Evaluation of Knowledge, Attitude and Performance of the mothers of Children Affected by Cutaneous Leishmaniasis. *Infectious Diseases: Research and Treatment*. 3:35–40: doi.org/10.4137/idrt. s3786.

Hendricks, L, Wright, N .1997. Diagnosis of Cutaneous Leishmaniasis by in Vitro Cultivation of Saline Aspirates in Schneider's Drosophila Medium, *The American Journal of Tropical Medicine and Hygiene*, 28 (6): 962 – 964. doi: https://doi.org/10.4269/ajtmh.1979.28.962.

Henten, S, Adriaensen, W, Fikre, H, Akuffo, H, Diro, E, Hailu, A, Van der Auwera, G & van Griensven, J. 2018. Cutaneous Leishmaniasis Due to Leishmania aethiopica. *EClinicalMedicine*. 6:69–81. doi.org/10.1016/j.eclinm.2018.12.009.

Hussein, NR, Balatay, AA, Saleem, ZSM, Hassan, SM, Assafi, MS, Sheikhan, RS, Amedi, FR, Hafzullah, SS, et al. 2019. A clinical study of cutaneous leishmaniasis in a new focus in the Kurdistan region, Iraq. *PLoS ONE*. 14(5):1–7. doi.org/10.1371/journal.pone.0217683.

Husson, F & Josse S. 2008. FactoMineR: An R package for multivariate analysis. *Journal of Statistical Software*. 25(1):1–18. doi.org/10.18637/jss. v025.i01.

Inceboz, T. 2019. Epidemiology and Ecology of Leishmaniasis. In: Rodriguez-Morales AJ, ed. *Current Topics in Neglected Tropical Diseases*. London, UK: IntechOpen Limited.

Jacquelyn, C. 2018. Leishmaniasis. Healthline Media a Red Ventures. https://www.healthline.com/health/leishmaniasis#prevention. (Accessed on Dec 26, 2020).

Jalayer, M & Zhou, H. 2016. A multiple correspondence analysis of at-fault motorcycle-involved crashes in Alabama. *Journal of Advanced Transportation*. 50(8):2089–2099. doi.org/10.1002/atr.1447.

JoeCaz .2006. Gojo Bet. Personal: Gojo Bet. joecaz.blogspot.com. (Accessed on Dec 26, 2020).

Jorjani, O, Mirkarimi, K, Charkazi, A, Shahamat, YD, Mehrbakhsh, Z & Bagheri, A. 2019. The epidemiology of cutaneous leishmaniasis in Golestan Province, Iran: A cross-sectional study of 8-years. *Parasite Epidemiology and Control*. 5(19): e00099. doi.org/10.1016/j.parepi. 2019.e00099.

Kamhawi, S. 2000. The biological and immunomodulatory properties of sand fly saliva and its role in the establishment of Leishmania infections. *Microb Infect*. 2(14):1765-73. https://doi.org/10.1016/S1286-4579(00)01331-9.

Karimi, A, Alborzi, A & Amanati, A. 2016. Visceral leishmaniasis: An update and literature review. *Archives of Pediatric Infectious Diseases*. 4 (3): e31612. doi.org/10.5812/pedinfect.31612.

Kassahun, A, Sadlova, J, Benda, P, Kostalova, T, Warburg, A, Hailu, A, Baneth, G, Volf, P, et al. 2015. Natural infection of bats with Leishmania in Ethiopia. *Acta Tropica*. 150:166–170. doi.org/10.1016/j.actatropica.2015.07.024.

Kassambara, A, and Fabian M. 2020. *Factoextra: Extract and Visualize the Results of Multivariate Data Analyses*. http://www.sthda.com/english/rpkgs/factoextra. (Accessed on 18 January 2020).

Kassi, M, Kassi, M, Afghan, AK, Rehman, R & Kasi, PM. 2008. Marring leishmaniasis: The stigmatization and the impact of cutaneous leishmaniasis in Pakistan and Afghanistan. *PLoS Neglected Tropical Diseases*. 2(10):1–3. doi.org/10.1371/journal.pntd.0000259.

Kebede, N, Worku, A, Ali, A, Animut, A, Negash, Y, Gebreyes, WA & Satoskar, A. 2016. Community knowledge, attitude, and practice towards cutaneous leishmaniasis endemic area Ochello, Gamo Gofa Zone, South Ethiopia. *Asian Pacific Journal of Tropical Biomedicine*. 6(7):562–567. doi.org/10.1016/j.apjtb.2016.01.018.

Kelbore, AG, Owiti, P, Reid, AJ, Bogino, EA, Wondewosen, L & Dessu, BK. 2019. Pattern of skin diseases in children attending a dermatology clinic in a referral hospital in Wolaita Sodo, southern Ethiopia. *BMC Dermatology*. 19(1):1–8. doi.org/10.1186/s12895-019-0085-5.

Khosravani Poor, H., Ghavam, A., Yazdan Panah, A., 2016. The Impact of Factors Related to Preventive Behaviors of Cutaneous Leishmaniasis Among Families of Kherameh 2015. *Pars Journal of Medical Sciences* (*Jahrom Medical Journal*). 14(1):63-70. Doi.org/10.29252/jmj.14.1.64.

Kirstein, OD, Faiman, R, Gebreselassie, A, Hailu, A, Gebre-Michael, T & Warburg, A. 2013. Attraction of Ethiopian phlebotomine sand flies (Diptera: Psychodidae) to light and sugar-yeast mixtures (CO§ssub§2§esub§). *Parasites and Vectors*. 6(1):1–9. doi.org/10.1186/1756-3305-6-341.

Lemma, W, Erenso, G, Gadisa, E, Balkew, M, Gebre-Michael, T & Hailu, A. 2009. A zoonotic focus of cutaneous leishmaniasis in Addis Ababa, Ethiopia. *Parasites and Vectors*. 2(1):1–8. doi.org/10.1186/1756-3305-2-60.

Lemma, W. 2018. Zoonotic leishmaniasis and control in Ethiopia. *Asian Pacific Journal of Tropical Medicine*. 11(5):313–319. doi.org/10.4103/1995-7645.233178.

Levin, KA. 2006. Study design III: Cross-sectional studies. *Evidence-Based Dentistry*. 7(1):24–25. doi.org/10.1038/sj.ebd.6400375.

Lima, LHGDM, Mesquita, MR, Skrip, L, De Souza Freitas, MT, Silva, VC, Kirstein, OD, Abassi, I, Warburg, A, et al. 2016. DNA barcode for the identification of the sand fly Lutzomyia longipalpis plant feeding preferences in a

tropical urban environment. *Scientific Reports*. 6:1–6. doi.org/10.1038/srep29742.

Linderman, E. 2011. "Procavia capensis" (On-line), Animal Diversity Web. https://animaldiversity.org/accounts/Procavia_capensis.(Accessed 19 November 2020).

Lindoso, JAL, Moreira, CHV, Cunha, MA, Queiroz, IT. 2018. Visceral leishmaniasis and HIV coinfection: current perspectives. *HIV AIDS (Auckl)*.;10:193-201. https://doi.org/10.2147/HIV.S143929.

Lobiondo-Wood, G & Harber, J. 2002. *Nursing research methods: critical appraisal and utilization. 5th edition.* St Louis: CV Mosby.

Luz, ZMP, da Silva, AR, Silva, F de O, Caligiorne, RB, Oliveira, E & Rabello, A. 2009. Lesion aspirate culture for the diagnosis and isolation of Leishmania spp. from patients with cutaneous leishmaniasis. *Memorias do Instituto Oswaldo Cruz*. 104(1):62–66. doi.org/10.1590/S0074-02762009000100010.

Maryam H, Mostafa HF, Leili A-M, Aliakbar M P, Sima R, Mehrnush S-N, Vahid Y, Farhad J-N & Mehdi Y. 2016. Recombinant Leishmaniamajor lipophosphoglycan 3 activates human Tlymphocytes via TLR2-independent pathway, *Journal of Immunotoxicology*, 13 (2):263-269, DOI: 10.3109/1547691X.2015.1066906.

Maza, MS & Oihane, M. 2014, Leishmaniasis transmission biology: Role of Promastigote SecretoryGel as a transmission determinant. PhD thesis, London School of Hygiene & Tropical Medicine. DOI: https://doi.org/10.17037/PUBS.01775854.

McCombes, S. 2019. *Research Design: Types, Methods, and Examples.* https://www.scribbr.com/methodology/research-design/ (Accessed on Febrewary 2,2019).

MedicineNet, Leishmaniasis (LeishmaniaInfection). 2007. https://www.medicinenet.com/leishmaniasis/article.htm (Accessed on March 23,2020).

Meles, TM. 2018. Species composition and habitat preference of sandflies in Ochollo village: a cutaneous leishmaniasis hotspot in Southwest

Ethiopia. MSc. THESIS. submitted to Arba Minch University, Ethiopia (obtained through personal communication).

Menezes, JPB, Guedes, CES, De Oliveira Almeida Petersen, AL, Fraga, DBM & Veras, PST. 2015. Advances in development of new treatment for leishmaniasis. *BioMed Research International*. (2015):15–18. doi.org/10.1155/2015/815023.

Mengesha, B, Endris, M, Takele, Y, Mekonnen, K, Tadesse, T, Feleke, A & Diro, E. 2014. Prevalence of malnutrition and associated risk factors among adult visceral leishmaniasis patients in Northwest Ethiopia: A cross-sectional study. *BMC Res Notes 7: 75. https://doi.org/10.1186/1756-0500-7-75.*

Ministry of Health -Ethiopia: Malaria Prevention & Control Program. 2020 http://www.moh.gov.et/ejcc/en/malaria-prevention-control-program (Accessed on October 23, 2020).

Mohammadiha, A, Dalimi, A, Mahmoodi, MR, Parian, M, Pirestani, M & Mohebali, M. 2017. The PCR-RFLP-based detection and identification of the Leishmania species causing human cutaneous leishmaniasis in the Khorasan-Razavi Province, Northeast of Iran. *Journal of Arthropod-Borne Diseases*. 11(3):383–392. PMCID: PMC5758634.

Mokobi F. 2020. Giemsa Stain- Principle, Procedure, Results, Interpretation. https://microbenotes.com/giemsa-stain-principle-procedure-results-interpretation/ (Accessed on Sep 18,2020).

Monde Medecins du. 2011. The KAP Survey Model (Knowledge, Attitudes, and Practices). https://www.spring-nutrition.org/publications/tool-summaries/kap-survey-model-knowledge-attitudes-and-practices. (Accessed on October 23, 2020).

Monroy-Ostria, A, Nasereddin, A, Victor, M. Monteon, Guzmán-Bracho, C & Jaffe C. 2014. ITS1 PCR-RFLP Diagnosis and Characterization of Leishmania in Clinical Samples and Strains from Cases of Human Cutaneous Leishmaniasis in States of the Mexican Southeast. *Interdisciplinary Perspectives on Infectious Diseases*. 2014 (6): 607287. http://dx.doi.org/10.1155/2014/607287.

Na-Bangchang, K, Ahmed, O, Hussein, J, Hirayama, K, Kongjam, P, Aseffa, A & Karbwang, J. 2016. Exploratory, Phase II Controlled Trial of Shiunko Ointment Local Application Twice a Day for 4 Weeks in Ethiopian Patients with Localized Cutaneous Leishmaniasis. *Evidence-based Complementary and Alternative Medicine*. 2016:5984709. doi: 10.1155/2016/5984709.

Nandha, B, Srinivasan, R & Jambulingam, P. 2014. Cutaneous leishmaniasis: Knowledge, attitude, and practices of the inhabitants of the Kani forest tribal settlements of Tiruvananthapuram district, Kerala, India. *Health Education Research*. 29(6):1049–1057. doi.org/10.1093/her/cyu064.

Nazari, M, Taravatmanesh, G, Kaveh, MH, Soltani, A & Ghaem, H. 2016. The effect of educational intervention on preventive behaviors towards cutaneous leishmaniasis at Kharameh city in 2014. *Shiraz E Medical Journal*. 17(10):4–10. doi.org/10.17795/semj39957.

Negera, E, Gadisa, E, Yamuah, L, Engers, H, Hussein, J, Kuru, T, Hailu, A, Gedamu, L, et al. 2008. Outbreak of cutaneous leishmaniasis in Silti woreda, Ethiopia: risk factor assessment and causative agent identification. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. 102(9):883–890. doi.org/10.1016/j.trstmh.2008.03.021.

Nikookar, SH, Pashaeei, T, Nikzad, D, Moosa-kazemi, SH, Davari, B, Control, V, Sciences, H, Branch, F, et al. 2015. Fauna and Larval Habitat Characteristics of Mosquitoes in Neka County, Northern Iran. *Journal of Arthropod-Borne Diseases*. 9:253-266. PMID: 30584547.

Nouiri, I, Chemak, F, Mansour, D, Bellalii, H, Ghrab, J, Baaboub, J & Chahed, MK. 2015. Impacts of irrigation water management on consumption indicators and exposure to the vector of Zoonotic Cutaneous Leishmaniasis (ZCL) in Sidi Bouzid, Tunisia. *Int. J. Agric. Pol. Res.* 3(2):93–103. http://dx.doi.org/10.15739/IJAPR.031.

Nuako KG. 2016. Investigation of a New Focus of Cutaneous Leishmaniasis in Ghana By. 1–236. https://eprints.lancs.ac.uk/id/eprint/79963/1/2016Kwakye_NuakoPhD.pdf . (Assessed on Dec 29, 2020).

Odonne, G, Berger, F, Stien, D, Grenand, P & Bourdy, G. 2011. Treatment of leishmaniasis in the Oyapock basin (French Guiana): A K.A.P. survey and analysis of the evolution of phytotherapy knowledge amongst Wayãpi Indians. *Journal of Ethnopharmacology*. 137(3):1228–1239. doi.org/10.1016/j.jep.2011.07.044.

Omari, H, Chahlaoui, A, Talbi, F, Ouarrak, K & El Ouali Lalami, A. 2020. Impact of Urbanization and Socioeconomic Factors on the Distribution of Cutaneous Leishmaniasis in the Center of Morocco. *Interdisciplinary Perspectives on Infectious Diseases*. 2020 :2196418. doi.org/10.1155/2020/2196418.

Oryan, A & Akbari, M. 2016. Worldwide risk factors in leishmaniasis. *Asian Pacific Journal of Tropical Medicine*. 9(10):925–932. doi.org/10.1016/j.apjtm.2016.06.021.

Oryan, A, Alidadi, A, Akbari, M. 2014. Risk Factors Associated with Leishmaniasis. *Tropical Medicine & Surgery*. 02(03):10–12. doi.org/10.4172/2329-9088.1000e118.

Özbilgin, A, Tünger, Ö, İnanır, I, Çavuş, İ, Perk, NE & Özel, Y. 2020. Comparison of liver extract medium with novy-macneal-nicolle medium and the molecular method for the diagnosis of leishmaniasis. *Klimik Dergisi*. 33(2):137–141. doi.org/10.5152/kd.2020.29.

Pardo, RH, Carvajal, A, Ferro, C & Davies, CR. 2006. Effect of knowledge and economic status on sandfly control activities by householders at risk of cutaneous leishmaniasis in the subandean region of Huila department, Colombia. *Biomédica: revista del Instituto Nacional de Salud*. 26 Suppl 1:167–179. doi.org/10.7705/biomedica.

Pareyn, M, Kochora, A, Van Rooy, L, Eligo, N, Broeckeid, B Vanden, Girma, N, Merdekios, B, Wegayehu, T, et al. 2020. Feeding behavior and activity of Phlebotomus pedifer and potential reservoir hosts of leishmania aethiopica in southwestern Ethiopia. *PLoS Neglected Tropical Diseases*. 14(3):1–19. doi.org/10.1371/journal.pntd.0007947.

Pareyn, M, Van den Bosch, E, Girma, N, van Houtte, N, Van Dongen, S, Van der Auwera, G, et al. 2019. Ecology and seasonality of sandflies and

potential reservoirs of cutaneous leishmaniasis in Ochollo, a hotspot in southern Ethiopia. PLoS Negl Trop Dis 13(8): e0007667. https://doi.org/10.1371/ journal. pntd.0007667.

Parvizi, MM, Zare, F, Handjani, F, Nimrouzi, M & Zarshenas, MM.2020. Overview of herbal and traditional remedies in the treatment of cutaneous leishmaniasis based on Traditional Persian Medicine. *Dermatologic Therapy*. 33(4): e13566. https://doi.org/10.1111/dth.13566.

Patiño-Londoño, SY, Salazar, LM, Acero, CT, Bernal, IDV. 2017. Socio-epidemiological and cultural aspects of cutaneous leishmaniasis: conceptions, attitudes, and practices in the populations of Tierralta and Valencia (Cordoba, Colombia). *Salud Colect* 13(1):123-138. doi:10.18294/sc..1079. https://pubmed.ncbi.nlm.nih.gov/28562730/

Pavli, A & Maltezou, HC. 2010. Leishmaniasis, an emerging infection in travelers. *International Journal of Infectious Diseases*. 14(12): e1032–e1039. doi.org/10.1016/j.ijid.2010.06.019.

Pedamkar P. 2020. Clustering Methods. https://www.educba.com/clustering-methods/ (Accessed on Dec 26, 2020).

Pigott, DM, Bhatt, S, Golding, N, Duda, KA, Battle, KE, Brady, OJ, Messina, JP, Balard, Y, et al. 2014. Global distribution maps of the leishmaniases. *eLife*. 3:1–21. doi.org/10.7554/elife.02851.

Polit, DF & Beck, CT. 2010. *Essentials of Nursing Research: Appraising evidence for nursing practice.* Seventh edition. Phiadelphia, Baltmore, Nework, London, Buenos Aires, Hong Kong, Sydney, Tokyo: Wolters Kluwer Health/Lippincott Williams & Wilkins.

Pourhoseingholi, M. A., Vahedi, M., & Rahimzadeh, M.2013. Sample size calculation in medical studies. *Gastroenterology and hepatology from bed to bench*, *6*(1), 14–17. PMC4017493.

Rakhshani, T, Kashfi, M, Ebrahimi, MR, et al. 2017. Knowledge, Attitude and Practice of the Households about Prevention of Cutaneous Leishmaniasis, Iran, Shiraz at 2016. *Journal of Human, Environment, and Health Promotion*. 2(3):186–192. doi.org/10.29252/jhehp.2.3.186.

Randa, M, Naguiba, F & Osama, M .2015. Attitude and knowledge of primary health care physicians and local inhabitants about leishmaniasis in west Alexandria. *Tropical Medicine and International Health*. 20(6):358. doi.org/10.5281/zenodo.1108813.

Rassi, Y, Saghafipour, A, Abai, MR, Oshaghi, M, Sina, R, Mehdi, M,Yaghoobi, E, Mohammad, R, Fatemeh, M & Babak, F. 2011. Phlebotomus papatasi and Meriones libycus as the vector and reservoir host of cutaneous leishmaniasis in Qomrood District, Qom Province, central Iran. *Asian Pacific journal of tropical medicine*. 4:97-100. 10.1016/S1995-7645(11)60045-X.

Rasti, S, Ghorbanzadeh, B, Kheirandish, F, Mousavi, SG, Pirozmand, A, Hooshyar, H & Abani, B. 2016. Comparison of Molecular, Microscopic, and Culture Methods for Diagnosis of Cutaneous Leishmaniasis. *Journal of Clinical Laboratory Analysis*. 30(5):610–615. doi.org/10.1002/jcla.21910.

Reithinger, R, Mohsen, M & Leslie, T. 2010. Risk factors for anthroponotic cutaneous leishmaniasis at the household level in Kabul, Afghanistan. *PLoS Neglected Tropical Diseases*. 4(3):1–8. doi.org/10.1371/journal.pntd.0000639.

Reveiz, L, Maia-Elkhoury, ANS, Nicholls, RS, Sierra Romero, GA & Yadon, ZE. 2013. Interventions for American Cutaneous and Mucocutaneous Leishmaniasis: A Systematic Review Update. *PLoS ONE*. 8(4): e61843. doi.org/10.1371/journal.pone.0061843.

Rich, RC, Brians, CL, Manheim, JB, Willnat, L . 2011. *Empirical Political Analysis: Quantitative and Qualitative Research Methods*. 8th ed. Boston, MA: Longman, 2011.

Richard JT. 2016. Writing Integrative Reviews of the Literature: Methods and Purposes. International Journal of Adult Vocational Education and Technology. 7(3):62-70. doi: 10.4018/IJAVET.2016070106.

Rohousova, I, Talmi-Frank, D, Kostalova, T, Polanska, N, Lestinova, T, Kassahun, A, Yasur-Landau, D, Maia, C, et al. 2015. Exposure to Leishmania spp. and sand flies in domestic animals in northwestern Ethiopia. *Parasites and Vectors*. 8(1) :360 doi.org/10.1186/s13071-015-0976-1.

Rojas, CA. 2001. An ecosystem approach to human health and the prevention of cutaneous leishmaniasis in Tumaco, Colombia. *Cadernos de saúde pública / Ministério da Saúde, Fundação Oswaldo Cruz, Escola Nacional de Saúde Pública*. 17 Suppl:193–200. doi.org/10.1590/s0102-311x2001000700029.

Roque, ALR & Jansen, AM. 2014. Wild and synanthropic reservoirs of Leishmania species in the Americas. *International Journal for Parasitology: Parasites and Wildlife*. 3(3):251–262. doi.org/10.1016/j.ijppaw.2014.08.004.

Ruoti, M, Oddone, R, Lampert, N, Orué, E, Miles, MA, Alexander, N, Rehman, AM, Njord, R, et al. 2013. Mucocutaneous leishmaniasis: Knowledge, attitudes, and practices among Paraguayan communities, patients, and health professionals. *Journal of Tropical Medicine*. 2013: 538629. doi.org/10.1155/2013/538629.

Rutte, EEA. 2018. *Visceral Leishmaniasis: Potential for Control and Elimination*. Erasmus University Rotterdam. Retrieved from http://hdl.handle.net/1765/103869. (Accessed on December 23, 2020).

Saberi, S, Zamani, A, Motamedi, N, Nilforoushzadeh, MA, Jaffary, F, Rahimi, E & Hejazi, SH. 2012. The knowledge, attitude, and prevention practices of students regarding cutaneous leishmaniasis in the hyperendemic region of the Shahid Babaie Airbase. *Vector-Borne and Zoonotic Diseases*. 12(4):306–309. doi.org/10.1089/vbz.2010.0259.

Saghafipour, A, Nejati, J, Mozaffari, E, Rezaei, F, Gharlipour, Z & Mirheydari, M. 2017. The effectiveness of education based on BASNEF model on promoting preventive behavior of cutaneous leishmaniasis in students. *International Journal of Pediatrics*. 5(6):5125–5136. doi.org/10.22038/ijp.2017.22373.1875.

Sakhaei, S, Darrudi, R, Motaarefi, H & Sadagheyani, HE. 2019. Epidemiological study of cutaneous leishmaniasis in Neyshabur county, East of Iran (2011-2017). *Open Access Macedonian Journal of Medical Sciences*. 7(21):3710–3715. doi.org/10.3889/oamjms.2019.421.

Saki, J, Akhlaghi, L, Maraghi, S, Meamar, AR, Mohebali, M, Oormazdi, H, Razmjou, E, Khademvatan, S, et al. 2009. Evaluation of modified Novy-MacNeal-

Nicolle medium for isolation of Leishmania parasites from cutaneous lesions of patients in Iran. *Research Journal of Parasitology*. 4(2):56–62. doi.org/10.3923/jp.2009.56.62.

Sambrook, J. & Russell, D. (2000). Molecular Cloning: a Laboratory Manual, 3rd edn. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory.

Sangiorgi, B, Miranda, DN, Oliveira, DF, Santos, EP, Gomes, FR, Santos, EO, Barral, A & Miranda, JC. 2012. Natural breeding places for phlebotomine sand flies (Diptera: Psychodidae) in a semiarid region of Bahia State, Brazil. *Journal of Tropical Medicine*. 2012: 124068. doi.org/10.1155/2012/124068.

Sara, KS. 2019. Leishmaniasis in Sweden. Molecular, diagnostic and epidemiological studies of the parasite Leishmania in a non-endemic country. Lund University: Faculty of Medicine. Doctoral Thesis (compilation). ISBNs 978-91-7619-753-0. https://portal.research.lu.se/en/publications/leishmaniasis-in-sweden-molecular-diagnostic-and-epidemiological (Accessed on December 23, 2020)

Sarkari, B, Qasem, A & Shafaf, MR. 2014. Knowledge, attitude, and practices related to cutaneous leishmaniasis in an endemic focus of cutaneous leishmaniasis, southern Iran. *Asian Pacific Journal of Tropical Biomedicine*. 4(7):566–569. doi.org/10.12980/APJTB.4.2014C744.

SastaSundar. 2020. Leishmaniasis. https://www.sastasundar.com/healtharticle/leishmaniasis (Accessed on December 23, 2020)

Scorza, BM, Carvalho, EM & Wilson, ME. 2017. Cutaneous manifestations of human and murine leishmaniasis. *International Journal of Molecular Sciences*. 18(6): 1296. doi.org/10.3390/ijms18061296.

Seid, A, Gadisa, E, Tsegaw, T, Abera, A, Teshome, A, Mulugeta, A, Herrero, M, Argaw, D, et al. 2014. Risk map for cutaneous leishmaniasis in Ethiopia based on environmental factors as revealed by geographical information systems and statistics. *Geospatial Health*. 8(2):377–387. doi.org/10.4081/gh.2014.27.

Seife, T, Benecha, AK, Zewdu, FT, Ayal, A & Misganaw, M. 2018. Treatment Patterns and Effectivness of Anti-Leishmaniasis Agents for Patients with Cutaneous Leishmaniasis at Boru Meda Hospital, South Wollo, Northeast Ethiopia, 2017/18. *Journal of Clinical & Experimental Dermatology Research*. 09(03):1–6. doi.org/10.4172/2155-9554.1000450.

Semeneh, G. 2012. The Psycho-Social Impact of Cutaneous Leishmaniasis on People Infected by the Disease.Masters desertation. Addis Ababa University. http://etd.aau.edu.et/.(assessed on January 3, 2019).

Serarslan, G, Ekiz, Ö, Özer, C & Sarıkaya, G. 2019. Dermoscopy in the Diagnosis of Cutaneous Leishmaniasis. *Dermatology Practical & Conceptual*. 9(2):111–118. doi.org/10.5826/dpc.0902a06.

Sheard, U. 2018. Quantitative data analysis, Publisher; Elsevier. Chapter18, Pages429-452, Number of pages24, Edition2nd, ISBN 9780081022214.

Shoshani, J. 2005. Order Hyracoidea. Pages 87-88 in Mammal Species of the World, 3rd Edition. D. E. Wilson and D. M. Reeder, eds. Johns Hopkins University Press.

Showler, A. J., Boggild, A. K. 2015. Cutaneous leishmaniasis in travellers: a focus on epidemiology and treatment in 2015. *Curr*. *Infect*. *Dis*. *Rep*.; 17: 37. https://doi.org/10.1007/s11908-015-0489-2.

Siriwardana, Y, Deepachandi, B, Gunasekara, C, Warnasooriya, W & Karunaweera, ND. 2019. Leishmania donovani induced cutaneous leishmaniasis: An insight into atypical clinical variants in sri lanka. *Journal of Tropical Medicine*. 2019:4538597. doi.org/10.1155/2019/4538597.

Sordo, L, Gadisa, E, Custodio, E, Cruz, I, Simón, F, Abraham, Z, Moreno, J, Aseffa, A, et al. 2012. Short report: Low prevalence of Leishmania infection in post-epidemic areas of Libo Kemkem, Ethiopia. *American Journal of Tropical Medicine and Hygiene*. 86(6):955–958. doi.org/10.4269/ajtmh.2012.11-0436.

Sorocco, T, Weigel, M, Armijos, R, Cevallos, W, Sanchez, X & Puebla, E. 2017. Cutaneous Leishmaniasis Knowledge, Attitudes and Practices (KAP)

Survey of an Endemic Rainforest Population in Northern Ecuador. *Annals of Global Health*. 83(1):165. doi.org/10.1016/j.aogh.2017.03.369.

Spronk, I, Korevaar, JC, Poos, R, Davids, R, Hilderink, H, Schellevis, FG, Verheij, RA & Nielen, MMJ. 2019. Calculating incidence rates and prevalence proportions: Not as simple as it seems. *BMC Public Health*. 19(1):1–9. doi.org/10.1186/s12889-019-6820-3.

Srinivasan, R, Ahmad, T, Raghavan, V, Kaushik, M & Pathak, R. 2018. Positive influence of behavior change communication on knowledge, attitudes, and practices for Visceral Leishmaniasis/Kala-Azar in India. *Global Health Science and Practice*. 6(1):192–209. doi.org/10.9745/GHSP-D-17-00087.

Stark, CG .2020. Leishmaniasis. Medscape. https://emedicine.medscape.com/article/220298-overview (assessed on January 3, 2019).

Stefan Ek. 2015. Gender differences in health information behaviour: a Finnish population-based survey. Health Promot Int. 2015 Sep;30(3):736-45. doi: 10.1093/heapro/dat063.

Steverding, D. 2017. The history of leishmaniasis. *Parasites and Vectors*. 10(1):1–10. doi.org/10.1186/s13071-017-2028-5.

Strassle, P, Hess, AS, Thom, KA, & Harris, AD. 2012. Assessing sensitivity and specificity in new diagnostic tests: the importance and challenges of study populations. *Infection control and hospital epidemiology*, *33*(11), 1177–1178. https://doi.org/10.1086/668036.

Sunter J, Gull K. 2017. Shape, form, function and Leishmania pathogenicity: from textbook descriptions to biological understanding. Open Biol. 7: 170165. http://dx.doi.org/10.1098/rsob.170165.

Suresh A. 2019. What is the Chi-Square Test and How Does it Work? An Intuitive Explanation with R Code . https://www.analyticsvidhya.com/blog/2019/11/what-is-chi-square-test-how-it-works/ (Accessed on Dec 26, 2020).

Swaminathan, S .2018. Logistic Regression — Detailed Overview. https://towardsdatascience.com/logistic-regression-detailed-overview-46c4da4303bc. (Accessed on December 23, 2020).

Taber, JM, Leyva, B & Persoskie, A. 2015. Why do People Avoid Medical Care? A Qualitative Study Using National Data. *Journal of General Internal Medicine*. 30(3):290–297. doi.org/10.1007/s11606-014-3089-1.

Tamiru, HF, Mashalla, YJ, Mohammed, R & Tshweneagae, GT. 2019. Cutaneous leishmaniasis a neglected tropical disease: Community knowledge, attitude and practices in an endemic area, Northwest Ethiopia. *BMC Infectious Diseases*. 19(1):1–10. doi.org/10.1186/s12879-019-4506-1.

Tankeshwar, A. 2016. NNN medium: Composition, Procedure and Results https://microbeonline.com/nnn-medium-composition-procedure-and-results/ (accessed on Sep 18,2020).

Tanure, A, Peixoto, JC, Afonso, MM dos S, Duarte, R, Pinheiro, A da C, Coelho, SVB & Barata, RA. 2015. Identification of Sandflies (Diptera: Psychodidae: Phlebotominae) Blood Meals in an Endemic Leishmaniasis Area in Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*. 57(4):321–324. doi.org/10.1590/s0036-46652015000400008.

Tareen, A, Afaq, S & Ul Haque, A. 2014. Comparative Study of the Diagnosis of Cutaneous Leishmaniasis by Slit Skin Smear and Skin Biopsy for Histopathology. *Journal of Rawalpindi Medical College (JRMC)*. 18(1):83–86.

Tasew, G, Kebede, A, Wolday, D, Gadisa, E, Britton, S, Eidsmo, L & Akuffo, H. 2009. Low-cost liquid medium for in vitro cultivation of Leishmania parasites in low-income countries. *Global Health Action*. 2(1):1–5. doi.org/10.3402/gha. v2i0.2046.

Tedla G, Bariagabr D, FH & Abreha, HH. 2018. Incidence and Trends of Leishmaniasis and Its Risk Factors in Humera, Western Tigray. *Journal of Parasitology Research*. 2018:8463097. doi.org/10.1155/2018/8463097.

Teklemariam, Z, Awoke, A, Dessie, Y & Weldegebreal, F. 2015. Ownership and utilization of insecticide-treated nets (ITNs) for malaria control in Harari

National Regional State, Eastern Ethiopia. *Pan African Medical Journal*. 21:1–9. doi.org/10.11604/pamj.2015.21.52.5380.

Thomas, M. 2018. Identification of Leishmania tropica and Leishmania aethiopica by DNA Sequencing and the Detection of Leishmania RNA Virus. Lancaster University. https://doi.org/10.17635/lancaster/thesis/200.

Tilahun, F, Alemu, W, Mulatu, G. 2014. Magnitude and Associated Factors of Cutaneous Leishmaniasis; in Mekelle City, Ayder Referral Hospital, Tigray, Northern Ethiopia, 2014. *Clinical Medicine Research*. 3 (6):189-199. doi: 10.11648/j.cmr.20140306.16

Torres-Guerrero, E, Quintanilla-Cedillo, MR, Ruiz-Esmenjaud, J & Arenas, R. 2017. Leishmaniasis: a review. *F1000Research*. 6(F1000 Faculty Rev):750. doi.org/https://doi.org/10.12688/f1000research.11120.1).

Vahabi, A, Rassi, Y, Oshaghi, MA, Vahabi, B, Rafizadeh, S & Sayyad, S. 2013. First survey on knowledge, attitude, and practice about cutaneous leishmaniasis among dwellers of Musian District, Dehloran County, Southwestern of Iran, 2011. *Life Science Journal*. 10(12 SPL.ISS.):864–868. ISSN:1097-8135.

Valderrama-Ardila, C, Alexander, N, Ferro, C, Cadena, H, Marín, D, Holford, TR, Munstermann, LE & Ocampo, CB. 2010. Environmental risk factors for the incidence of American cutaneous leishmaniasis in a sub-andean zone of Colombia (Chaparral, Tolima). *American Journal of Tropical Medicine and Hygiene*. 82(2):243–250. doi.org/10.4269/ajtmh.2010.09-0218.

Vivero, RJ, Torres-Gutierrez, C, Bejarano, EE, Peña, HC, Estrada, LG, Florez, F, Ortega, E, Aparicio, Y, et al. 2015. Study on natural breeding sites of sand flies (Diptera: Phlebotominae) in areas of Leishmania transmission in Colombia. *Parasites and Vectors*. 8(1):1–14. doi.org/10.1186/s13071-015-0711-y.

Von Davier, M & Lee, Y. 2019. Handbook of Diagnostic Classification Models Models and Model Extensions, Applications, Software Packages: 10.1007/978-3-030-05584-4. (Accessed on 18 January 2020).

Votýpka, J, Kasap, OE, Volf, P, Kodym, P & Alten, B. 2012. Risk factors for cutaneous leishmaniasis in Cukurova region, Turkey. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. 106(3):186–190. doi.org/10.1016/j.trstmh.2011.12.004.

Vries de, Reedijk, HJC, SH & Schallig, HDFH. 2015. Cutaneous Leishmaniasis: Recent Developments in Diagnosis and Management. *American Journal of Clinical Dermatology*. 16(2):99–109. doi.org/10.1007/s40257-015-0114-z.

Wall, EC, Watson, J, Armstrong, M, Chiodini, PL & Lockwood, DN. 2012. Short report: Epidemiology of imported cutaneous leishmaniasis at the hospital for tropical diseases, London, United Kingdom: Use of polymerase chain reaction to identify the species. *American Journal of Tropical Medicine and Hygiene*. 86(1):115–118. doi.org/10.4269/ajtmh.2012.10-0558.

Weerakoon, HS, Ranawaka, RR, Bandara, WMP, Herath, P & Warnasekara, YPJN. 2016. Knowledge on Leishmaniasis among Health Care Workers in Endemic Area. *Anuradhapura Medical Journal*. 10(1):6. doi.org/10.4038/amj. v10i1.7597.

Wijerathna, T, Gunathilaka, N, Gunawardena, K & Rodrigo, W. 2020. Socioeconomic, demographic and landscape factors associated with cutaneous leishmaniasis in Kurunegala District, Sri Lanka. *Parasites and Vectors*. 13(1):1–14. doi.org/10.1186/s13071-020-04122-1.

Wijesinghe, H, Gunathilaka, N, Semege, S, Pathirana, N, Manamperi, N, De Silva, C & Fernando, D. 2020. Histopathology of Cutaneous Leishmaniasis Caused by Leishmania donovani in Sri Lanka. *BioMed Research International*. 2020:4926819. doi.org/10.1155/2020/4926819.

Winston-Salem State University. 2020. Pre-Award - Key Elements of a Research Proposal Quantitative Design https://www.wssu.edu/...ice-of-sponsored-programs/pre-award/ - 36KB - 2020-08-15 02:03:51. (assessed on 2 February 2020).

World Health Organisation. 2014. Regional Strategic Framework for Elimination of Kala-azar from the South-East Asia. *Sea-Cd-239*. 24. http://apps.searo.who.int/pds_docs/B4870.pdf?ua=1%0Ahttp://www.sear

o.who.int/entity/world_health_day/2014/KA_CD239.pdf. (Accessed on 2 February 2019).

World Health Organization. 2010. Control of the leishmaniases. WHO technical report series 949. Report of a meeting of the WHO expert committee on the control of the leishmaniases Geneva. https://apps.who.int/iris/bitstream/handle/10665/44412/WHO_TRS_949_eng.pdf;sequence=1. (Accessed on February 2,2019).

World Health Organization. 2020. Leishmaniasis. https://www.who.int/news-room/fact sheets/detail/leishmaniasis. (Accessed 3 October 2019).

World Health Organization. 2020. Neglected tropical diseases. Cutaneous leishmaniasis factsheet. http://www.emro.who.int/neglected-tropical-diseases/information resources-leishmaniasis/cl-factsheet.html.(Accessed on 29 Oct 2020).

World Health Organizathion.2018. World Health Organization. 2018. Available from: https://www.who.int/leishmaniasis/burden/GHO_VL_2018.pdf?ua=1. (Accessed 3 October 2019).

World Health Organizathion.2020. Available from http://www.who.int/leishmaniasis/resources/ECUADOR.pdf. (Accessed on 13 July 2020).

World Health Organization.2020. https://www.who.int/leishmaniasis/burden/Leishmaniasis_Colombia/en/. www.who.int/leishmaniasis/resources/COLOMBIA.pdf.(Accessed 3 October 2019).

World Health Organization .2012. Regional strategic framework for elimination of kala-azar from the South-East Asia Region (2011-2015). Regional Office for South-East Asia WHO Regional Office for South-East Asia. https://apps.who.int/iris/handle/10665/205826. (Accessed on 19 June 2019).

World Health Organization .2016. Leishmaniasis fact sheet. https://www.who.int/news-room/fact-sheets/detail/leishmaniasis (Accessed on 2 February 2019).

World Health Organization. 2012. The Post Kala-azar Dermal Leishmaniasis (PKDL) Atlas. A manual for health workers. 1–216. doi.org/10.1016/S0033-3182(99)71269-7. (Accessed on 19 June 2019).

World Health Organization. 2018. Leishmaniasis; http://www.who.int/news-room/fact-sheets/detail/leishmaniasis. (Accessed 31 March 2018).

World Health Organization. 2020. Global leishmaniasis surveillance, 2017–2018, and first report on 5 additional indicators. From https://www.who.int/publications/i/item/who-wer9525 (Accessed 18 August 2020).

World Health Organization. 2020. Health statistics and information systems. About the Global Burden of Disease (GBD) project. https://www.who.int/healthinfo/global_burden_disease/about/en/ (Accessed on 23 December 2020).

Yohannes, M, Abebe, Z & Boelee, E. 2019. Prevalence and environmental determinants of cutaneous leishmaniasis in rural communities in Tigray, northern Ethiopia. *PLoS Neglected Tropical Diseases*. 13(9): e0007722. doi.org/10.1371/journal.pntd.0007722.

Younis, LG, Kroeger, A, Joshi, AB, Das, ML, Omer, M, Singhid, VK, Gurung, CK & Banjara, MR. 2020. Housing structure including the surrounding environment as a risk factor for visceral leishmaniasis transmission in Nepal. *PLoS Neglected Tropical Diseases*. 14(3):1–13. doi.org/10.1371/journal.pntd.0008132.

Zijlstra, EE. 2014. PKDL and Other Dermal Lesions in HIV Co-infected Patients with Leishmaniasis: Review of Clinical Presentation in Relation to Immune Responses. *PLoS Neglected Tropical Diseases*. 8(11): e0003258. doi.org/10.1371/journal.pntd.0003258.

Zolfaghari F, Khosravi H, Shahriyari A, Jabbari M, Abolhasani A. 2019. Hierarchical cluster analysis to identify the homogeneous desertification management units. PLoS ONE 14(12): e0226355. https://doi.org/10.1371/journal.pone.0226355.

Annexure A: KAP survey questioner (English version)

KAP SURVEY QUESTIONNAIRE

Survey objective: To explore CL- related knowledge, attitudes and practice of persons bitten by suspected rabid animals

Check one: Date: ___ / ___ / ___

Baseline data collection; or Sub city: _____ site: _____

Follow-up data collection code: _____

Information to read to respondent:

We'd like to learn more about your CL knowledge, attitudes, and practices. We intend to identify your requirements and the most effective manner to deliver information to you, as well as any barriers to getting medical care. CL control will be improved with the information you supply. Your responses will be kept private and will not be shared with anybody. Your name will not appear on the questionnaire or be recorded in any other way. Your participation in the interview is entirely voluntary, and you may terminate it at any time.
Thank you for your assistance.

Interviewer: Place an X in the box of the selected answer(s). Do not read responses unless the directions indicate.

A. DEMOGRAPHIC INFORMATION

1. How old are you? _____ Years**.**

1. □ 15-24
2. □ 25–34
3. □35- 44
4. □ 45-54
□Over 55

2. What is your gender? (Record by observing)

1. □Male
2. □Female

3. What is the highest level of education you have completed?

1. □ No formal school
2. □ Elementary or junior
3. □High school (9-12grade)
4. □College
5. □Higher education (professional or post-graduate)
6. □Religious schooling only

4. Duration of stay at residence _____
5. Household number _____
6. Place of birth _____
7. If migrant; Place of origin _____ year of arrival _____
8. Occupation _____

1. □ Agricultural labor
2. □ Services
3. □ Skilled laborer
4. □Business
5. □government employee
7. □ Other (specify): _____

8.1. If your income source is farming, the location of the farmland is

1= near home
2= near the river/_____ (in Butajirra)
3 = both

9. Religion _____

1. □ Orthodox
2. □ Muslim
3. □Protestant
4. □catholic
5. □ Pagan
6. □ Other (specify): _____

B. GENERAL KNOWLEDGE OF CL

10. Ask the respondent if they could name the disease after showing a picture of CL manifestation

1. □ Able to identify as CL 2. □ Unable to identify
3. □ other: _____

11. Have you heard about Cutaneous Leishmaniasis (Chewi)?1. □yes 2. □No

12. If your answer is 'yes" to the question above, how did you hear about CL? (Please choose the three most effective sources.)

1. □ Newspapers and magazines 2. □TV 3. □Radio
4. □Billboards 5. □Health workers 6. □ Religious leaders
7. □Brochures, posters and other printed materials 8. □Teachers
9. □Family, friends, neighbors and colleagues
10. □Other (please explain): _____

13. What is the cause of CL?

1.□spiritual or hereditary 2. □ contact with patient
3. □virus 4. □Leishmanaia
5.□Mosquito bite 6. □the bite of sandfly
7. □Germ 8. □microbe
9. □I do not know
10. □other: _____

14. What are the signs of CL?

1. □ fever with chills 3. □Plaque
2. □Skin rash 4. □Papule
5. □ Ulcer 6. □I do not know
7. □other: _____

15. Where in the body are lesions/scars of CL Located?

1. □ Forehead 2. □ Face 3. □ Nostril
4. □ Arm 5. □ Leg 6. □ Ear
8. □ Mixed 7. □I do not know
9. □ other: _____

16. Is CL an infectious disease/ can be transmitted from one person to another?

1. □yes 2. □No 3. □I do not know

17. Is there Possibility of acquiring leishmaniasis in travelling to endemic areas?

1. □yes 2. □no 3. □I do not know

18. Is complete cure from CL possible?

1. □yes 2. □no 3. □I do not know

19. Ask the respondent if they know about the biting and blood sucking behavior after showing live sand flies collected from their environment:

1. □ yes, I know 2. □ No, I don't know

20. Where do you think is the breeding place of the vector?

1. □ Dirty place 4. □Damp and dark places
2. □ Cervices in the house 5. □ water ponds
3. □ Thatched roof 6. □ Garbage collection sites
7. □Cattle sheds
8. □ do not know
9. □Others: _____

21. When is the preferred biting time of the vector?

1. □Dusk 3. □Midnight

2. □anytime whether day or night4. □ I do not know

C. THE ATTITUDES TOWARDS CL

22. Do you think CL can be treated?
1. □ yes 2. □ no 3. □ I have no idea

24. Do you believe that the occurrence of CL in one member of the family affects the economy of the whole family?
1. □ yes 2. □ no 3. □ I have no idea

25. What do you think the outcome of CL if not treated?
1. □ death 2. □ disability 3. □ self cure
4. □ I have no idea 5. □Other: _____

26. Do you believe that environmental health is important for prevention of transmission?
1. □ yes 2. □ no 3. □ I have no idea

27. What do you think are the major constraints to control CL?
1. □an insufficient budget 3. □ Religious taboo
2. □ Trained professionals 4. □ Lack of appropriate legislation
5. □Awareness 6. □Lack of proper coordination
7. □ I do not know
8. □Others: _____

28. What is yours preferred drug of choice for treatment of CL?
1. □ Specific medicine 2. □Indigenous medicine
3. □ Do not know
4. □other: _____

28. If you would not go to the health facility, what is the reason? (Please check all that apply.)
1. □ Not sure where to go 1. □Yes 0. □No
2. □Cost 1. □Yes 0. □No
3. □ Difficulties with transportation/distance to clinic 1. □Yes 0. □No
4. □Do not trust medical workers 1. □Yes 0. □No
5. □Do not like attitude of medical workers 1. □Yes 0. □No
6. □Cannot leave work 1. □Yes 0. □No
7. □Don't want to find out that something is wrong 1. □Yes 0. □No
8. □ preference to use herbal medication 1. □Yes 0. □No
9. □Other (please explain): _____

29. How expensive do you think CL diagnosis and treatment is in this country?
1. □ It is free of charge 2. □It is reasonably priced
3. □ It is somewhat/moderately expensive 4. □It is very expensive
5. □I have no idea

30. Do you feel well informed about CL?
1. □Yes 2. □ No

31. If your answer to the above question is "no" whose mistake is that?
1. □Your negligence
2. □ You never had the chance to learn about rabies
3. □there is inadequate source of information in your locality
4. □other: _____

32. Have you ever participated in CL control activities?
1. □Yes 2. □No (reason: _____)

33. How serious a problem do you think CL is in your locality? (Check one.)
    1. □Very serious 2. □Ordinary 3. □Not very serious
    4. □I don't know

D. PRACTICES REGARDING CL

34. How can someone with CL be treated? (Check all that are mentioned.)

1. □Herbal remedies	1. □Yes	0. □No
2. □ Home rest without medicine	1. □Yes	0. □No
3. □Praying	1. □Yes	0. □No
4. □ Specific drugs given by health centre	1. □Yes	0. □No
5. □ vaccination	1. □Yes	0. □No
6. □holy water	1. □Yes	0. □No

    7. □Do not know
    8. □Other: _____

35. Do you think CL is preventable?
    1. □ yes 2. □ no 3. □ I have no idea

36. What are the preventive/control measures against CL?

1. □ Using insecticides	1. □Yes	0. □No
2. □ Early diagnosis and treatments	1. □Yes	0. □ No
3. □ Using covered clothes	1. □Yes	0. □No
4. □ Use of bed nets	1. □Yes	0. □No
5. □ Use of insect repellents	1. □Yes	0. □No
6. □ Reduce outdoor activities	1. □Yes	0. □No
7. □ avoiding contact with animals	1. □Yes	0. □No
8. □ cleanliness	1. □Yes	0. □No

    9. □covering of the ulcers
    10. □Do not know
    11. □Other: _____

37. Do you have bed net in the house?
    1. □yes (how many: _____) 2. □ no

38. Do you have custom of spending time outside in the night or sleeping outdoor?
    1. □ yes 2. □ no

39. Do you use bed nets when sleeping? 1. □ yes 2. □ no

40. When is your Work time preference when the temperature is high?
    1.□Day time 2. □Night 3. □Both

41. Do you use repellents? 1. □yes 2. □ no

42. Do you properly perform garbage disposal
    1. □ yes (how often; _____) 2. □ no

43. Has your house ever been sprayed?
    1. □ yes (how often; _____)
    2. □ no (reason: _____)

Thank you very much for participating in our survey.

Annexure B: KAP survey questioner (Amharic version)

Knowledge attitude and practice *ቃለ-መጠይቅ*

Survey objective: *ይህ ቃለ-መጠይቅ የተዘጋጀዉ ጨቄ የሚባለዉን በሽታ በተመለከተ የወረዳዉን ነዋሪዎች ዕዉቀት ዝንባሌና ተግባራዊ ልምድ ለመገምገም ነዉ::*

Check one: ቀን: ___ / ___ / ___

ወረዳ: _____ ቀበሌ: _____

ጎጥ;_____

ኮድ:_____ ስም:- _____ ስልክ

ቁጥር:_____

ለተጠያቂዉ የሚነበብ:
የርስዎን በጨቄ በሽታ ላይ ያሎትን ዕዉቀት ዝንባሌና ተግባራዊ ልምድ ለማወቅ እንፈልጋለን፤ ይህም የርስዎን ፍላጎት ለመረዳትና የተሻለ የጤና እንክብካቤ ለማምጣት ይረዳናል፡፡ በተጨማሪም እርሶ የሚሰጡን መረጃ የጨቄን በሽታ ለመቆጣጠር ይረዳናል፡፡ የእርሶ መልስ በምንም መልኩ ለሌላ አገልግሎት ዐይዉልም፡፡በዚህ መጠይቅ ላይ የሚሳትፉት ሙሉ ፍቃደኛ ሲሆኑ ብቻ ነዉ፡፡ በፈለጉት ጊዜም መጠይቁን ማቁዋረጥ ይችላሉ፡፡
በጥናቱ ላይ ስለተሳትፉ በጣም እናመሰግናልን፡፡

ለጠያቂዉ: **የተመረጠዉ መልስ(መልሶች) ላይ በሳጥኑ ዉስጥ** X ምልክት ያስቀምጡ.

በትዛዝ ካልተጠቀሰ በቀር ምርጫዎችን አያንብቡ.

B. DEMOGRAPHIC INFORMATION

1. *ዕድሜዎ?* _____ *ዓመት.*

3. □ 18.0-24.5 3. □34.5- 44.5
4. □ 24.5–34.5 4. □ 44.5-54.5 □ ከ 54.5 ዓመት በላይ

2. ጸታ? (በመመልከት ይመዝግቡ) 1. □ወንድ 2. □ ሴት

3. *ሀይማኖት* _____

1. □ አርቶዶክስ 4. □ ካቶሊክ
2. □ ሙስሊም 5. □ ሌላ (ይጥቀሱ): _____
3. □ፕሮቴስታንት

4. የትምህርት ደረጃዎ?

1. □ ማንበብና መጻፍ የማይችሉ 4. □ ሁለተኛ ደረጃ(7-12grade)
2. □ማንበብና መጻፍ የሚችሉ 5. □ ኮሌጅ/ከፍተኛ ትምህርት
3. □ የመጀመሪያ ደረጃ (1-6) 6. □ ሌላ (ይጥቀሱ): _____

5.በዚህ ቀበሌ የሮሩበት ግዜ (በዓመት) _____

6. የቤተሰብ መጠን _____

7. የትዉልድ ስፍራ _____

8. ከሌላ ቦታ ከመጡ; የመጡበት ቦታ (የከተማዉ መጠሪያ) _____ወደዚህ የመጡበት ዓመት_____

9. ስራ _____

1. □ ገበሬ 2. □ አገልግሎት ሰጭ
3. □ ነጋዴ 4. □ የቤት እመቤት
5. □የመንግስት ሰራተኛ 7. □ ሌላ (ይጥቀሱ): _____

10. ዋና የገቢ ምንጭዎ የእርሻ ስራ ከሆነ, እርሻዎ የሚገኘዉ

1.□ ከቤት አጠገብ 2. □ ከወንዝ አጠገብ/_____ 3. □ ሁለቱም ጋር

B. GENERAL KNOWLEDGE OF CHEWIE

11. ስህል ቁጥር 1 ካሳዩ በኋላ ተጠያቂዉ የበሽታዉን መጠሪያ ያዉቁ እንደሆን ይጠይቁአቸዉ

1. □ ጨቄ ብለዉ ለይተዋል 2. □ መለየት አልቻሉም

12.ስለ ጨቄ በሽታ ሰምተዉ ያዉቃሉ? 1. □ አዎ 2. □አይ

13. ከላይ ላለዉ ጥያቄ መልስዎ አዎ ከሆነ, ስለበሽታዉ እንዴት ሰሙ? (የተጠቀሱት ላይ ምልክት ያድርጉ)

1. □ ጋዜጣና መጽሄት 1. □ አዎ 2. □አይ
2.□ቴሌቪዥን 1. □ አዎ 2. □አይ
3.□ራድዮ 1. □ አዎ 2. □አይ
4. □ማስታወቂያ 1. □ አዎ 2. □አይ
5.□የጤና ባለሙያዎች 1. □ አዎ 2. □አይ
6.□ የሀይማይኖት አለቆች 1. □ አዎ 2. □አይ
7. □በራሪ ወረቀቶች እና ሌሎች ህትመቶች 1. □ አዎ 2. □አይ
8.□መምራኖች 1. □ አዎ 2. □አይ
9. □ቤተሰብ፤ጓደኛ እና ጎረቤቶች 1. □ አዎ 2. □አይ
10. □ሌላ (ይጥቀሱ): _____

13.1. በአካባቢዎ በጨቄ በሽታ የተያዘ ሰዉ ያዉቃሉ?
1. □ አዎ (አሁን የተያዘ__□__/ከዚህ በፊት የተያዘ__□__) 2. □አይ

13.2. ከላይ ላለዉ ጥያቄ መልስዎ አዎ ከሆነ የተያዘዉ ሰዉ ማን ነበር/ነዉ?
1. □የቤተሰብ አባል 2. □ እራሴ
3. □ጎረቤት 4. □የሩቅ ዘመድ 5. □ ሌላ(ይጥቀሱ): _____

14. ጨቄ በሽታ ከምን የሚመጣ ይመስልዎታል? (ከአንድ በላይ መልስ መስጠት ይቻላል)
1.□መንፈሳዊ 2. □ ከዘር የሚመጣ 3. □ሌይሽማንያ
4. □ቫይረስ 5. □ተዋሲያን 6. □ጀርም
7. □አላዉቅም 8. □ሌላ (ይጥቀሱ): _____

15. ጨቄ በሽታ እንዴት የሚተላለፍ ይመስልዎታል?
1.□ በወባ ትንኝ ንክሻ 2. □ከታመመ ሰዉ ጋር ባለ መነካካት
3. □ የወባ ትንኝ መሳይ ግን አነስ ባለች ፍጥረት 4. □ ቆሻሻ ዉሃ በመጠጣት
5.□ የተበላሸ ምግብ በመመገብ 6. □ አላዉቅም
7. □ሌላ (ይጥቀሱ): _____

16. የጨቄ በሽታ ምልክቶቹ ወይም ስሜቶቹ ምንድን ናቸዉ? (ከአንድ በላይ መልስ መስጠት ይቻላል)
1. □ ትኩሳት 2. □ቀላ ብሎ ያበጠ (Plaque) 3. □የቆዳ መቆጣት
4. □ቁንጥር ቁስል/መምገል 5. □እትሬ (ቆዳ ላይ ያተረ)-papule 6. □አላዉቅም
7. □ሌላ (ይጥቀሱ): _____

17. የትኛዉ የሰዉነት ክፍላችን በብዛት በበሽታዉ ይጠቃል? (ከአንድ በላይ መልስ መስጠት ይቻላል)
1. □ ግንባር 2. □ ፊት 3. □ አፍንጫ 4. □ እጅ
5.□ እግር 6. □ ጆሮ 7. □ ከአንድ ቦታ በላይ 8. □ አላዉቅም
9. □ሌላ (ይጥቀሱ): _____

18. ጨቄ ካንዱ ሰዉ ወደ ሌላ የሚተላለፍ በሽታ ነዉ ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

19. በሽታዉ በብዛት የተሰራጨበት ቦታ የተጓዘ ሰዉ በበሽታዉ የመያዝ እድሉ ይጨምራል ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

20. ከታከሙ በኋላ ሙሉ በሙሉ መዳን ይቻላል ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

21. ስዕል ቁጥር 2 በማሳየት ስለትንኟ የንክሻ እና ደም የመምጠጥ ፀባይ ተጠያቂዉ ያዉቁ እንደዉ ይጠይቁአቸዉ:
1. □ አዎ አዉቃለሁ 2. □ አይ አላዉቅም

22. ትንኟ የምትራባበት ቦታ የት ይመስልዎታል?
1. □ ቆሻሻ ቦታ 4. □ ጨለማ ቦታ
2. □ ቤት ዉስጥ በሚገኙ ስንጥቅጥቅ ቦታዎች 5. □ ዉሃ ያቆረ ቦታ
3. □ የሳር ክዳን ባለዉ ጣሪያ 6. □ ቆሻሻ ማጠራቀሚያ ቦታ
7. □የከብቶች ማረፊያ 8. □ አላዉቅም
9. □ሌላ (ይጥቀሱ):_____

23.ትንኟ ለመንከስ የምትመርጠዉ ግዜ የቱ ነዉ?

3. □አመሻሽ ላይ 3. □እኩለ ሌሊት ላይ
4. □ በማንኛዉም ጊዜ 4. □ አላዉቅም

C. THE ATTITUDES TOWARDS CHEWIE

24. ጨቄ መታከም የሚችል በሽታ ነዉ ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

25. በአንድ ቤተሰብ አባል ዉስጥ ጨቄ በሽታ ቢከሰት የቤተሰቡን ኢኮኖሚ ይጎዳል ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

26. ጨቄ በሽታ ካልታከመ ምን የሚያመጣ ይመስሎታል?
1. □ ሞት 2. □ አካል ጉዳት 3. □ እራሱ ይድናል
4. □ አላዉቅም 5. □ ሌላ (ይጥቀሱ): _____

27. የአካብቢ ንፅህና ጨቄን ለመከላከል ይጠቅማል ብለዉ ያስባሉ?
1. □አዎ 2. □አይደለም 3. □አላዉቅም

28. ጨቄን ለመቆጣጠር እንቅፋት ይሆናሉ ብለዉ የሚጠቅሱአቸዉ ነገሮች (ከአንድ በላይ መልስ መስጠት ይቻላል)
1. □የበጀት እጥረት 3. □ የተሳሳቱ አመለካከቶች
2. □ የጤና ባለሙያዎች እጥረት 4. □ ተገቢ ህግ አለመኖር
5. □የግንዛቤ እጥረት 6. □ቅንጅት አለመኖር
7. □ አላዉቅም 8. □ሌላ (ይጥቀሱ): _____

29. ጨቄን ለማከም የመጀመሪያ ምርጫዎ ምንድን ነዉ?
1. □ ዘመናዊ ሕክምና 2. □ባህላዊ ሕክምና
3. □ አላዉቅም 4. □ሌላ (ይጥቀሱ): _____

30. ወደ ዘመናዊ ሕክምና ካልሄዱ, ምክንያቱ ምንድን ነዉ? (ከአንድ በላይ መልስ መስጠት ይቻላል)

A. □ ወዴት መሄድ እንዳለብኝ አለማወቅ	1. □ አዎ	2. □አይ
B. □ወጭ መብዛቱ	1. □ አዎ	2. □አይ
C. □ መጓጓዣ ችግር	1. □ አዎ	2. □አይ
D. □ዘመናዊ ሕክምና ስለማያድን	1. □ አዎ	2. □አይ
E. □የሕክምና ባለሙያዎች በስርዓት ስለማያስተናግዱ	1. □ አዎ	2. □አይ
F. □ስራ ትቼ መሄድ ስለማልችል	1. □ አዎ	2. □አይ
G. □በምርመራ በሽታ እንደተገኘብኝ ማወቅ ስለማልፈልግ	1. □ አዎ	2. □አይ
H. □ የባሕል ሕክምናን ስለምመርጥ	1. □ አዎ	2. □አይ

I. □ሌላ (ይጥቀሱ):_____

31. በአካባቢዎ ባለዉ ሕክምና ቦታ ለጨቄ ምርመራ የሚያወጡት ወጭ እንዴት ነዉ?
1. □ ክፍያ የለዉም 2. □ተገቢ ክፍያ ነዉ
3. □ ትንሽ ወደድ ይላል 4. □በጣም ዉድ ነዉ
5. □አላዉቅም

32. በአካባቢዎ ባለዉ ሕክምና ቦታ ለጨቄ ሕክምና የሚያወጡት ወጭ እንዴት ነዉ?
1. □ ክፍያ የለዉም 2. □ተገቢ ክፍያ ነዉ
3. □ ትንሽ ወደድ ይላል 4. □በጣም ዉድ ነዉ
5. □አላዉቅም

33. ስለ ጨቄ በቂ መረጃ አለኝ ብለዉ ያስባሉ?
1. □አዎ 2. □ አይ

34. ከላይ ላለዉ ጥያቄ መልስዎ አይ ከሆን የማን ክፍተት ነዉ?
1. □የራሴ ቸልተኝነት 2. □ ስለብሽታዉ የመማር እድል አላገኘሁም
3.□በአካባቢዬበቂመረጃየለም 4. □ሌላ(ይጥቀሱ): _____

35. በአካባቢዎ የጨቄ በሽታ መቆጣጠሪያ ተደርጎ ቢሆን እርስዎ ተሳትፈዉ ያዉቃሉ/መሳተፍ ይፈልጋሉ?
1. □አዎ 2. □አይ (ምክንያቱ: _____)

36. በአካባቢዎ ጨቄ ምን ያህል ጉዳት አምጭ ነዉ?

1. □በጣም ጎጂ 2. □መካከለኛ 3. □ጉዳቱ ያን ያህል አይደለም
4. □አላዉቅም

D. PRACTICES REGARDING CHEWIE

37. በጨቄ የተያዘ ሰዉ እንዴት ይታከማል? (ከአንድ በላይ መልስ መስጠት ይቻላል)

1. □በባሕል ሕክምና	1. □ አዎ	2. □አይ
2. □ ሳይታከሙ ቤት በማረፍ	1. □ አዎ	2. □አይ
3. □በፀሎት	1. □ አዎ	2. □አይ
4. □ከጤና ጣቢያ በሚሰጥ መድሃኒት	1. □ አዎ	2. □አይ
5. □ በክትባት	1. □ አዎ	2. □አይ
6. □በፀበል	1. □ አዎ	2. □አይ
7. □አላዉቅም	8. □ሌላ(ይጠቀስ): _____	

38. የጨቄን በሽታ መከላከል ይቻላል ብለዉ ያስባሉ?

1. □አዎ 2. □አላስብም 3. □አላዉቅም

39. ጨቄን የመከላከያ እና የመቆጣጠሪያ መንገዶች ምን ምን ናቸዉ?

1. □ ፀረ-ተባይ በመጠቀም	1. □ አዎ	2. □አይ
2. □ በጊዜ ተመርምሮ በመታከም	1. □ አዎ	2. □አይ
3. □ አቡጀዲ በመጠቀም	1. □ አዎ	2. □አይ
4. □ አጎበር በመጠቀም	1. □ አዎ	2. □አይ
5. □ እበትና የተለያዩ ቅጠሎችን በመታጠን	1. □ አዎ	2. □አይ
6. □ ከቤት ዉጭ ያሉ እንቅስቃሴዎችን በመቀነስ	1. □ አዎ	2. □አይ
7. □ ከእንስሳቶች ጋር ንክኪ ማስወገድ	1. □ አዎ	2. □አይ
8. □ የአካባቢን ንጽህና በመጠመቅ	1. □ አዎ	2. □አይ
9. □ቁስልን በመሸፈን	1. □ አዎ	2. □አይ
10. □አላዉቅም	11. □ሌላ(ይጠቀስ): _____	

40. ቤት ዉስጥ አጎበር አለዎት? 1. □አዎ (ምን ያህል: _____) 2. □ የለኝም

41. በማታ ዉጭ የማሳለፍ ወይም ከቤት ዉጭ የማደር ልምድ አሎት?

1. □ አዎ 2. □ የለኝም

42. ሲተኙ ብዙ ጊዜ አጎበር ይጠቀማሉ? 1. □ አዎ 2. □ አልጠቀምም

43. ሙቀት በሚጨምርበት ሰአታ በማሳዎ ላይ መቼ መስራት ይመርጣሉ?

1.□በቀን 2. □ማታ 3. □በቀንም በማታም

44. የትንኝ ማባረሪያ ይጠቀማሉ? 1. □ አዎ 2. □ አልጠቀምም

45. ቆሻሻ በምን ያህል ጊዜ ያስወግዳሉ?

1. □ በየቀኑ 2.□ በየሳምንቱ
3. □በየአስራ አምስት ቀኑ 4. □በየወሩ
5. □ ሌላ(ይጠቀስ) :_____

46. ቤትዎ ፀረ-ተባይ ተረጭቶ ያዉቃል?

1. □ አዎ (ምን ያህል ጊዜ; _____)
2. □ አይ (ለምን ምክንያት: _____)

47. ቤትዎ ተረጭቶ ከሆነ ለምን ተብሎ ነዉ የተረጨዉ

በጥናቱ ላይ ስለተሳተፎ ብጣም እናመሰገናለን!!!

ስም ፊርማ

ጠያቂ: _____ _____

አስተባባሪ: አቶ ሰብስቤ ከበደ _____

ተመራማሪ፡ ዶ/ር ሊና ጋዙ _____

ሱፐርቫይዘር፡ ዶ/ር ንጋቱ ከበደ _____

Annexure C: Risk factor assessment survey questioner (English Version)

Survey objective: To explore CL- To explore different factors affecting CL transmission in Sodo Wereda Community in SNNP, Ethiopia

Check one: Date: ___ / ___ / ___

Baseline data collection; or Wereda: _____Keble: _____

Follow-up data collection code: _____

Information to read to respondent:

We'd like to learn more about the risk factors that put this population at risk for CL." We'd like to learn more about your needs and the best way to get information to you, as well as any obstacles to getting medical help. CL control will be improved with the information you supply. Your responses will be kept private and will not be shared with anybody. Your name will not appear on the questionnaire or be recorded in any other way. Your participation is entirely voluntary, and you have the option to terminate the interview at any time.

Thank you for your assistance.

Interviewer: Place an X in the box of the selected answer(s).
Do not read responses unless the directions indicate.

a. DEMOGRAPHIC INFORMATION

1. How old are you? _____ Years**.**

5. □ 18.0-24.5
6. □ 24.5–34.5
3. □34.5- 44.5
4. □ 44.5-54.5
□Over 54.5

2. What is your gender? (Record by observing)

2. □Male
2. □Female

3. Religion _____

1. □ Orthodox
2. □ Muslim
3. □Protestant
4. □Catholic
5. □ Pagan
6. □ Other (specify): _____

4. What is the highest level of education you have completed?

1. □ No formal school
2. □ Elementary or junior
3. □High school (9-12grade)
4. □College
5. □Higher education (professional or post-graduate)
6. □Religious schooling only

5. Duration of stay at residence (In years) _____
6. Family size _____
7. Place of birth of respondent _____
8. If migrant; Place of origin (Town) _____ year of arrival _____
9. Occupation _____

1. □ Farmer
2. □ Services provider
3. □ Trader
4. □ Housewife
5. □government employee
7. □ Other (specify):_____

10. If your main income source is farming, the location of the farmland is

1.□ near home
2.□ near the river/_____
3.□ both

CL status

12. Infecte with CL?
    1. □ yes 2. □ no
13. if your answer is Yes to the above question, when was the disease aquired?
    1. □ currently 2. □ in the past 3. □ Other (specify):----------

If current infection	**If past infection**
14. when did you notice the disease symptom? ------- day --------- month ---------- year	18. if it was past infection, who was infected? 1. I 2. Other family member
15. Are you treated? 1. □ Yes (when did you get it terated -------day --------- month ---------- year) 2. □ No	19. when in the past were you infected? ------- Day --------- month ---------- year 20. is there a scar (observation)? 1. □ Yes 2. □ no 21. Where in the body id the scar located (Obseravtion)? Specify :_____
16. where did you get treatment? 1. □ Traditional (specify :----------) 2. □ Modern medication (Was the disease confirmed? 1. □ Yes 2. □ no)	22. How did you get it treated in the past? 1. □Use of herbal medicines 2. □Treatment at health facility 3. □Healed by itself 4. □Other (Specify :----------)
17. Is another member of the family with CL? 1. □ Yes 2. □ No	23. When did it heal? ------- Day --------- Month ---------- year

24. Geographical coordinates

Latitude:	^{0}Degree	Minutes (‘)
longitude:	^{0}Degree	Minutes (‘)
Attitude:		m.a.s.l

25. information on family members (to be filled by all members of the family)

R.No	Age (In years)	Gender 1. □Male 2. □Female	Education	CL status 1. □Positive 2. □Negative If positive 1. □Active lesion 2. □Scars
1.				
2.				

3.				
4.				
5.				
6.				
7.				
8.				
9.				

Educational status 1. □Illitrate 2. □ Able to read and write 3. □ Primary (1-6) 4. □ secondary (7-12) 5. □College/higher 6. □ Other (specify)：

_____

ENVIRONMENTAL AND HOST FACTORS FOR TRANSMISSION OF CL

A. HOUSEHOLD FACTOR

1. What is the type of wall surface?
1. □ mud 2. □bricks
3. □ stone 4. □wood
5. □ Other_____

2. Condition of the wall (filled by the interviewer observation)
1. □Cracked 3. □No Crack or holes
2. □Holes formed 4. □Only few areas

3. If cracked**,**
1.□ almost all walls 2. □ some of the walls

4. What is the type of ceilings/roof?
1. □cloth 3. □ Concrete
2. □ wood 4. □Straw
5. □Other_____

5. Type of floor
1. □cemented 3. □ pasted with cow dung/mud
2. □covered with straw
4. □other: _____

6. Is the latrine functional? 1.□ present 2. □ absent

7. If latrine is present, the condition is (filled by interviewer observation)
1. □ Good 2. □ Fair 3. □bad

8. Is the House located close to creeks or waterways? 1. □ Yes 2. □ No

9. Is there a Pit near house? 1. □ Yes 2. □ No

10. Are there screens on the windows and doors in your home?
1. □Yes 2. □Partial 3. □No

11. If yes, are they intact and in good condition? 1. □ Yes 2. □ No

12. How much is your monthly income?
1. □< 500 birr 3. □1000-2000 birr
2. □500- 1000 birr 4. □2000-4000 birr
5. □>4000 birr

13. How much of your earnings are you going to spend for food in Dry season?
1.□ <10% 2. □10-20%
3. □31-50% 4. □ 21-30%
5. □ >50%

14. Do you think every member of the family is well nourished (both in amount and kind)? 1.□ yes 2. □ No

B. ENVIRONMENTAL AND ECOLOGICAL FACTORS

15. Do you have a garden or a field in your residence?
1. □Yes 2. □No 3. □Don't know

16. Is there a public park near your home?
1. □Yes 2. □No 3. □Don't know

17. Farm present within 300 meters radius (approximate) 1. □ Yes 2. □ No

18. Types of trees /bushes in the garden/ compound (list the most common once only)
1. _____
2_____
3_____
4 _____
5_____
___

19. Are there any cave or gorges_____?
A. near your house 1. □ Yes 2. □ No
B. in your village 1. □Yes 2.□No
C. near your working area 1. □ Yes 2. □ No
D. other; _____

20. If yes, How far the cave/gorge from your house/working place? (Meters)
_____

21. Is your village expanding into previously abandoned forests? 1. □ Yes 2. □ No

22. 19- What kind of water supply does your residence have?
1. □Mains 2. □Vehicle
3. □ Well 4. □Others, please specify.....……

C. HOST FACTORS

23. What is the condition of Waste disposal around house? (Answer by observation)
1.□ open 2. □ closed

24. Do you keep Dry wood stored inside the house? 1. □ Yes 2. □ No

25. Where and how do you sleep?
1. □ Mat/Mattress with bed net. 2. □Mat/Mattress without a bed net.
3. □Under the tree shade (floor) 4. □traditional tower (*kote*)
5. Others (specify)_____

26. Do you have practice of spending near/at Gorge Early morning or nighttime?
1. □ yes 2. □ No

27. If is yes, how long do you spend near/at gorge minutes
_____

28. If is yes, why do you spend time near or at gorge?
_____

29. Have you or some of your family number visited other areas where CL is prevalent?
1. □ Yes 2. □ No

30. Do you sit in the home yard in the evenings? 1. □ Yes 2. □ No

31. If your answer is yes, how often?
1. □ always 3. □ Sometimes
2. □ frequently 4. □ Rarely

32. Do you sleep in the home yard? 1. □ Yes 2. □ No
33. Do you usually use impregnated bed nets? 1. □ Yes 2. □ No
34. Do you sleep your face/hands covered? 1. □ Yes 2. □ No
35. Do you have habit of working in the garden early morning? 1. □ Yes2. □ No
36. Do you have habit of irrigating crops at night? 1. □ Yes 2. □ No
37. Do you have custom of Living in temporary rural shelters (for farming, hunting or lumbering)?
1.□ yes 2. □ No
38. Do children play in the forest or in the night? 1. □ Yes 2. □ No
39. Do you use personal protective measures against mosquito, sandfly, or other insect bites?
1.□ yes 2. □ No
40. If yes to the question above which ones
A. Wear appropriate clotting (long sleeved shirts, long pants, boots, hats) 1. □ Yes 2. □ No
B. Use of bed nets 1. □ Yes 2. □ No
C. Apply lotion/repellent on the skin 1. □ Yes 2. □ No
D. Treat clotting with repellents 1. □ Yes 2. □ No
E. Other: _____
41. When do you prefer to work in the farm areas when temperature peaks?
1. □Day time 2. □In the night 3. □ both at night& daytime

D. ANIMAL FACTORS

42. Have you seen hyraxes living _____?
A. near your house 1. □ Yes 2. □ No
B. in your village 1. □Yes 2. □No
C. near your working area 1. □ Yes 2. □ No
D. other; _____
43. Have you seen bats living_____?
A. near your house 1. □ Yes 2. □ No
B. in your village 1. □Yes 2. □No
C. near your working area 1. □ Yes 2. □ No
D. other;
_____
44. You think that the population of hyrax/bats is increasing in your area?
1. □ Yes 2. □ No
45. If there are hyraxes near your house, do they come to your compound?
1. □ Yes 2. □ No
46. Are Domestic animals kept near/in house? 1.□ Yes 2. □ No
47. If yes which animals
1. □Cow 3. □Horses
2. □Donkey 4. □Sheep and goat
5. □ others: _____
48. Do you have cattle?
1. □ Yes 2. □ No
49. If yes to question above, where do you keep them,

1. □inside the house
2. □nearby animal house
3. □far from the house
4. □other, please specify.....

50. Pet animals kept near/in house? 1.□ Yes 2. □ No

51. If yes which pet animals?

1. □Dog 2. □Cat 3. □Others: _____

52. Do you have habit of damping animal dung near house? 1. □ Yes 2. □ No

53. Is there Presence of animal burrow around house (approximately within 300mtrs.radius)?

1.□ yes 2. □ No

Thank you very much for participating in our survey

Annexure D: Risk factor assessment survey questioner (Amharic Version)

ቃለ-መጠይቅ

Survey objective: ይህ ቃለ-መጠይቅ የተዘጋጀዉ ጨዌ የሚባለዉን በሽታ በተመለከተ የወረዳዉን ነዋሪዎች ዕዉቀት ዝንባሌና ተግባራዊ ልምድ ለመገምገም ነዉ::

Check one: ቀን: ___ / ___ / ___

ወረዳ: _____ **ቀበሌ:** _____

ጎጥ;_____

ኮድ:_____ **ስም:-** _____ **ስልክ**

ቁጥር:_____

ለተጠያቂዉ የሚነበብ

የርስዎን በጨዌ በሽታ ላይ ያሎትን ልምድ ለማወቅ እንፈልጋለን፤ ይህም የርስዎን ፍላጎት ለመረዳትና የተሻለ የጤና እንክብካቤ ለማምጣት ይረዳናል፡፡ በተጨማሪም እርሶ የሚሰጡን መረጃ የጨዌን በሽታ ለመቆጣጠር ይረዳናል፡፡ የእርሶ መልስ በምንም መልኩ ለሌላ አገልግሎት ዐይዉልም፡፡በዚህ መጠይቅ ላይ የሚሳትፉት ሙሉ ፍቃደኛ ሲሆኑ ብቻ ነዉ፡፡ በፈለጉት ጊዜም መጠይቁን ማቁዋረጥ ይችላሉ፡፡

በጥናቱ ላይ ስለተሳትፉ በጣም እናመሰግናልን፡፡

ለጠያቂዉ: የተመረጠዉ መልስ(መልሶች) ላይ በሳጥኑ ዉስጥ X ምልክት ያስቀምጡ፤በትዛዝ ካልተጠቀሰ በቀር ምርጫዎችን አያንብቡ.

C. DEMOGRAPHIC INFORMATION

1. ዕድሜዎ? _____ ዓመት.

2. ጾታ? (በመመልከት ይመዝግቡ) 1. □ወንድ 2. □ ሴት

3. ሀይማኖት _____

1. □ ኦርቶዶክስ
2. □ ሙስሊም
3. □ፕሮቴስታንት
4. □ ካቶሊክ
5. □ ሌላ (ይጥቀሱ): _____

4. የትምህርት ደረጃዎ?

1. □ ማንበብና መጻፍ የማይችሉ
2. □ማንበብና መጻፍ የሚችሉ
3. □ የመጀመሪያ ደረጃ (1-6)
4. □ ሁለተኛ ደረጃ(7-12grade)
5. □ ኮሌጅ/ከፍተኛ ትምህርት
6. □ ሌላ (ይጥቀሱ): _____

5.ዉልደትዎ እዚሁ ወረዳ ነዉ? 1. □ አዎ 2. □ አይ

6. ከሌላ ቦታ ከመጡ; የመጡበት ቦታ (የከተማዉ መጠሪያ) _____ወደዚህ የመጡበት ዓመት_____

7. የቤተሰብ መጠን _____

8. ስራ _____ 1. □ ገበሬ 2. □ ሌላ

9.1. ስራዎ ከግብርና ሌላ ከሆነ እባክዎን ይጥቀሱ

1. □ አገልግሎት ሰጭ
2. □ ነጋዴ
3. □ የቤት እመቤት
4. □የመንግስት ሰራተኛ
5. □ ሌላ (ይጥቀሱ): _____

10. ዋና የገቢ ምንጭዎ የእርሻ ስራ ከሆነ, እርሻዎ የሚገኘዉ

1.□ ከቤት አጠገብ 2. □ ከወንዝ አጠገብ/_____ 3. □ ሁለቱም ጋር

11. የጋብቻ ሁኔታ

1. □ ያገባ/ያገባች
2. ያላገባ/ያላገባች

CL status/ የጨዌሽታ ሁኔታ

12. በጨዌ በሽታ ተይዘዋል?

1. □ አዎ 2. □ አይ

13. ለጥያቄ 11 መልስዎ አዎ ከሆነ በበሽታዉ የተያዙት?

1. □አሁን ነዉ 2. □ከዚህ በፊት 3. □ሌላ(ይጥቀሱ)፡----------

በበሽታዉ የተያዙት አሁን ከሆነ	በበሽታዉ የተያዙት ከዚህ በፊት ከሆነ
14. የበሽታዉን ምልክት ያዩበት የመጀመሪያ ጊዜ መቼ ነበር? ------- ቀን --------- ወር---------- አ.ም	18. ከዚህ በፊት የተያዙ ከነበረ በበሽታዉ የተያዙት? 3. እርስዎ 4. ሌላ የቤተሰብ አባል
15. ህክምና አድርገዋል? 1. □ አዎ (ህክምና ያደረጉት መቼ ነዉ ------- ቀን --------- ወር---------- አ.ም) 2. □ አይ	19. ከዚህ በፊት የተያዙ ከነበረ መቼ ነበር? ------- ቀን --------- ወር---------- አ.ም 20. ሰዉነትዎ ላይ ከጨዉ የተነሳ ጠባሳ አለ? 1. □ አዎ 2. □ አይ 21. ከላይ ላለዉ ጥያቄ መልስዎ አዎ ከሆነ ጠባሳዉ የትኛዉ የሰዉነት ክፍል ላይ ነዉ? ባከዎን ይጥቀሱ፡ _____
16. ህክምና ካደረጉ የት ነዉ ያደረጉት? 1. □ ባህላዊ ህክምና (ይጥቀሱ፡----------) 2. □ ዘመናዊ ህክምና (ምርመራ ዉጤቱ በሽታዉ እንዳለቦት ተረጋግጦ ነበር 1. □ አዎ 2. □ አይ)	22. ከዚህ በፊት የተያዙ ከነበረ እንዴት ጠፋሎት? 5. □ባህላዊ ህክምና በማድረግ 6. □ወደ ጤናቋም በመሄድና በመታከም 7. □እራሱ ጠፋ 8. □ሌላ(ይጥቀሱ፡----------)
17. በበሽታዉ የተያዘ ሌላ የቤተሰብ አባል አለ? 1. □ አዎ 2. □ አይ	23. ከዚህ በፊት የተያዙ ከነበረ በሽታዉ መቼ ጠፋሎት? ------- ቀን --------- ወር---------- አ.ም

24. Geographical coordinates

Latitude:	^{0}Degree	Minutes (‘)
longitude:	^{0}Degree	Minutes (‘)
Attitude:		m.a.s.l

25. ስለሌሎች ቤተሰብ አባል መረጃ(to be filled by all members of the family)

ተራ.ቁ	ዕድሜ (በአመት)	ጾታ 3. □ወንድ 4. □ሴት	የትምህርት ደረጃ	CL status 1. □Positive 2. □Negative If positive 1. □Active lesion 2. □Scars
10.				
11.				
12.				
13.				

14.				
15.				
16.				
17.				
18.				

የትምህርትደረጃ 1. □ማንበብና መጻፍ የማይችሉ 2. □ ማንበብና መጻፍ የሚችሉ 3. □ የመጀመሪያ ደረጃ (1-6) 4. □ሁለተኛ ደረጃ (7-12) 5. □ኮሌጅ/ከፍተኛ ትምህርት 6. □ሌላ(ይጥቀሱ)፡_____

C.ENVIRONMENTAL AND HOST FACTORS

A. HOUSEHOLD FACTOR

54. የቤቱ ግድግዳ ምን አይነት ነዉ? (በመመልከት ይመዝግቡ)

1. □ ጭቃ/እበት 2. □ሸክላ 3. □ ድንጋይ
4. □እንጨት 5. □ ሌላ(ይጠቀስ): _____

55. የቤቱ ግድግዳ ሁኔታ (በመመልከት ይመዝግቡ)

1. □የተሰነጠቀ 3. □የተሰነጠቀም ሆነ ቀዳዳ የሌለዉ
2. □ ቀዳዳ ያለበት 4. □የተሰነጠቀ እና ቀዳዳ ያለበት
5. ሌላ(ይጠቀስ): _____

56. የተሰነጠቀ ከሆነ, 1. □ ከሞላ ጎደል ሁሉም ግድግዳ 2. □ የተወሰነዉ ግድግዳ

57. የቤቱ ጣሪያ ምን አይነት ነዉ? (በመመልከት ይመዝግቡ)

1. □ጨርቅ 3. □ ኮንክሪት 5. □ ቆርቆሮ
2. □ እንጨት 4. □ገለባ 6. □ሌላ(ይጠቀስ): _____

58. የቤቱ ወለል አይነት? (በመመልከት ይመዝግቡ)

3. □ሲሚንቶ 3. □ በእበት ወይም በጭቃ የተለቀለቀ
2.□በገለባ የተሸፈነ 4. □ሌላ(ይጠቀስ): _____

59. አገልግሎት እየሰጠ ያለ ሽንት ቤት አለ? 1.□ አለ 2. □ የለም

60. ሽንት ቤቱ ካለ, ሁኔታዉ ምን ይመስላል? (በመመልከት ይመዝግቡ)

2. □ ጥሩ 2. □ በቂ 3. □መጥፎ/ጥሩ ያልሆነ

61. ቤትዎ ከምንጭ ወይም ዉሃ ከሚወርድበት አጠገብ ነዉ? 1. □ አዎ 2. □ አይደለም

62. ቤትዎ አጠገብ የተቆፈረ ጉድጓድ አለ? 1.□ አለ 2. □ የለም

63. ቤትዎ በር ወይም መስኮት ላይ የትንኝ መከላከያ(screen) አለ? 1.□ አለ 2. □ የለም

64. አዎ ካሉ የሚሰራ እና በጥሩ ሁኔታ ላይ ያለ ነዉ? 1. □ አዎ 2. □ አይደለም

65. የወር ገቢዎ ምን ያህል ይገመታል

3. □< 500 ብር 3. □1000-2000 ብር
4. □500- 1000 ብር 4. □2000-4000 ብር
6. □>4000 ብር 6. ሌላ(ይጠቀስ): _____

66. በበጋ ወቅት ምን ያህሉን ገቢዎን ለምግብ ያዉላሉ?

1.□ <10% 2. □10-20%
3. □31-50% 4. □ 21-30%
5. □ >50% 6. ሌላ(ይጠቀስ): _____

67. የቤተሰብዎ አባላት ሁሉ በጥሩ ሁኔታ ምግብ እያገኙ ይመስልዎታል (በምግቡ መጠንም ሆነ አይነት መሆኑን ያስረዱ)?

1.□ አዎ 2. □ አይመስለኝም

D. ENVIRONMENTAL AND ECOLOGICAL FACTORS

68. በመሮሪያዎ ጓሮ ወይም ሜዳ አለ? 1.□አለ 2. □የለም

69. ቤትዎ አቅራቢያ የህዝብ መሰብሰቢያ/ሜዳ አለ? 1.□አለ 2. □የለ

70. ከቤትዎ 300 ሜትር ዙሪያ የእርሻ ቦታ አለ (በግምት)? 1.□አለ 2. □የለም

71. የዛፍ አይነቶች /በጓሮ/ በግቢ እና በአካባቢ (በጣም በብዛት የሚገኙትን በማስቀደም ይጠቀስ)

1. የግራር 1. □አለ 2. □የለም
2. ሰንሰል 1. □አለ 2. □የለም
3. ሌላ(ይጥቀሱ)_____

72. ዋሻ ወይም ገደላማ ሸለቆ አለ_____?

A. ቤትዎ አካባቢ 1. □አለ 2. □የለም
B. በመንደርዎ 1. □አለ 2. □የለም
C. በስራዎ አካባቢ 1. □አለ 2. □የለም
D. □ሌላ(ይጠቀስ):_____

20. አለ ካሉ, ዋሻዉ ወይም ገደላማዉ ሸለቆ ከቤትዎ/ከስራዎ ቦታ ምነ ያህል ይርቃል? (ግምት በሜትር)

_____

21. መንደርዎ በፊት ወደ ተተወ ጫካ እየሰፋ ነዉ? 1. □ አዎ 2. □ አይደለም

22. በአካባቢዎ ያለዉ የዉሃ ምንጭ ምንድን ነዉ?

2. □ወንዝ ዉሃ 2. □የቧንቧ ዉሃ
3. □ ጉድጓድ ዉሃ 4. □ሌላ(ይጠቀስ): _____

23. ከወባ ዉጭ በሌላ ትንኝ ተነክሰዉ ያዉቃሉ

1. □ አዎ 2. □ አይ 3. □ አላዉቅም

E. HOST FACTORS

24. በቤቱ አካባቢ ያለዉ የቆሻሻ አወጋገድ ምን ይመስላል? (ተመልከተዉ መዝግቡ)

1.□ ክፍት 2. □ የተዘጋ

25. በቤትዎም ሆነ በደጅዎ ደረቅ እንጨት ያከማቻሉ? 1. □ አዎ 2. □ አይ

26. የት እና እንዴት ይተኛሉ?

1. □ አጎበር ያለዉ ፍራሽ ላይ 2. □አጎበር የሌለዉ ፍራሽ ላይ
3. □በዛፍ ጥላ ስር (ወለል ላይ) 4. □ቆጥ ላይ
5. □ መሬት ላይ 6. □ሌላ(ይጠቀስ): _____

27. በማለዳ ወይም ማምሻዉን ገደላማ ሸለቆዉ አካባቢ ጊዜ የማሳለፍ ልምድ አለዎት?

2. □ አለኝ 2. □ የለኝም

28. አዎ ካሉ, ምን ያህል ጊዜ ነዉ ገደላማ ሸለቆዉ አካባቢ የሚያሳልፎት (ግምት በደቂቃ)

_____

29. አዎ ካሉ, በገደላማ ሸለቆዉ አካባቢ ጊዜ የሚያሳልፎት ለምንድን ነዉ?

_____

30. እርስዎ ወይም የቤተሰብዎ አባል ባለፉት 12 ወራት ወደ ሌላ አካባቢ ተጉዘዋል?

1. □ አዎ(የተጓዙበትን ቦታ ይጥቀሱ፡----------) 2. □ አይ

31. ቤትዎ ደጅ ላይ በማታ ይቀመጣሉ? 1. □ አዎ 2. □ አይ

32. መልስዎ አዎ ከሆነ, ምን ያህል ጊዜ?

1. □ ሁልጊዜ 3. □ አንዳንዴ
2. □ በተደጋጋሚ 4. □ በጣም አልፎ አልፎ

33. ደጅዎ ወይም ጊቢዎ ላይ ይተኛሉ? 1. □ አዎ 2. □ አይ

34. ብዙ ጊዜ ፀረ-ተባይ የተረጨ አጎበር ይጠቀማሉ? 1. □ አዎ 2. □ አይ

35. ሲተኙ ፊትዎን እና እጅዎን ይሸፍናሉ? 1. □ አዎ 2. □ አይ

36. በጓሮዎ ዉስጥ በማለዳ የመስራት ልምድ አለዎት? 1. □ አዎ 2. □ አይ

37. ሰብልዎን በማታ መስኖ የማጠጣት ልምድ አለዎት? 1. □ አዎ 2. □ አይ

38. በጊዜአዊ የገጠር መጠለያዎች(ዳስ) ዉስጥ የመሮር ልምድ አለዎት (ለእርሻ ስራ, ለአደን ወይም ለሌላ ጉዳይ)?

1. □ አዎ 2. □ አይ

39. ልጆች ምሽት ላይ ጫካ ዉስጥ ይጫወታሉ? 1. □ አዎ 2. □ አይ

40. እራስዎን ከወባ ትንኝ፤ ከሳንድ ትንኝ ወይም ከሌሎች የትንኞት ንክሻ የመከላከያ ዘዴዎችን ይጠቀማሉ?

1. □ አዎ 2. □ አይ

41. ከላይ ላለዉ ጥያቄ መልስዎ አዎ ከሆነ የትኛዉን ዘዴ?

F. ተገቢ ልብስ (እጅጌ ረጅም ሹራብ, ረጅም ሱሪ, ቡትስ ጫማ, ኮፍያ) 1. □ አዎ 2. □ አይ
G. አጎበር በመጠቀም 1. □ አዎ 2. □ አይ
H. ቅባት/ትንኝ ማባረሪያ መቀባት 1. □ አዎ 2. □ አይ
I. ልብስን በትንኝ ማባረሪያ በማድረግ 1. □ አዎ 2. □ አይ
J. □ሌላ(ይጠቀስ): _____

42. ሙቀቱ ከፍተኛ በሚሆንበት ወቅት ምን ሰዓት ነዉ እርሻዎ ላይ መስራት የሚፈልጉት?

1. □በቀን ሰዓት 3. □ በቀንም በማታም
2. □በማታ 4. □በማለዳ 5. □ሌላ(ይጠቀስ): _____

F. ANIMAL FACTORS

43. ሽኮኮ የሚኖሩ አሉ _____?

A. ቤትዎ አካባቢ 1. □አለ 2. □የለም
B. በመንደርዎ 1. □አለ 2. □የለም
C. በስራዎ አካባቢ 1. □አለ 2. □የለም
D. □ሌላ(ይጠቀስ): _____

44. የሌሊት ወፎች የሚኖሩ አሉ _____?

A. ቤትዎ አካባቢ 1. □አለ 2. □የለም
B. በመንደርዎ 1. □አለ 2. □የለም
C. በስራዎ አካባቢ 1. □አለ 2. □የለም
D. □ሌላ(ይጠቀስ): _____

45. በአካባቢዎ የሽኮኮ ወይም የሌሊት ወፎች ቁጥር እየጨመረ ይመስልዎታል?

1. □ አዎ 2. □ አይ

46. ቤትዎ አካባቢ ሽኮኮዎች ካሉ,ወደ እርስዎ ቅጥር ይመጣሉ? 1.□ አዎ 2. □ አይ

47. የቤት እንሰሶች በቤት ዉስጥ/አካባቢ ይቀመጣሉ? 1.□ አዎ 2. □ አይ

48. መልስዎ አዎ ከሆነ፣ የትኞቹ?

3. □ላሞች 3. □ፈረስ
4. □አህያ 4. □በግና ፍየል 5. □ ሌላ(ይጠቀስ): _____

49. መልስዎ አዎ ከሆነ, የት ይቀመጣሉ?

5. □መኖሪያ ቤት ዉስጥ 3. □ከመኖሪያዉ ቤት አካባቢ ሆኖ ግን በራሳቸዉ ቤት
6. □ከመኖሪያ ቤት እርቆ 4. □ሌላ(ይጠቀስ): _____

50. ዉሻና ድመት ቤትዎ አለ? መልስዎ አዎ ከሆነ የትኞቹን? 1.□ዉሻ 2. □ድመት

የከብቶችን እበት ቤትዎ አካባቢ ያጠራቅማሉ? 1. □ አዎ 2. □ አይ

51. የትናንሽ እንስሳቶች መደበቂያ ጉድጓድ በቤትዎ አካባቢ አለ (በግምት 300 ሜትር ዙሪያ ከቤትዎ)?

1.□ አዎ 2. □ አይ

52. በአካባቢዎ ያለዉ ጤና ተቋም ለእርስዎ የተመቸ ነዉ? 1.□ አዎ 2. □ አይ

53. ከላይ ላለዉ ጥያቄ መልስዎ አይ ከዎነ ምክንያቱ ምንድን ነዉ?

_____

በጥናቱ ላይ ስለተሳተፎ በጣም እናመሰገናለን!!

ስም ፊርማ

ጠያቂ: _____ _____

Annexure E: Clinical data collection sheet

Epidemiological investigation of Cutaneous Leishmanaisis in Sodo district
Sample collection sheet

Date: _____/_____/_____
Code: _____
Kebele:

- Kela Zuria ☐ Michael Semero ☐ Beka ☐
- Kola Nurena ☐ Genete Mariam ☐ Adazer ☐
- Other _____

Patient's personal detail
Age: _____
Sex: _____
Clinical and laboratory information

1. Date of appearance of first signs of the disease: __/__/____
2. Date of definitive diagnosis: __/__/____
3. No of lesions:
   1=one ☐ 2=two ☐ 3 = three or more ☐
4. Duration of lesion/s since first appeared in ☐☐☐ months
5. Site of lesion
   - 5.1. Cheek 1= yes_____ 2= no_____
   - 5.2. If yes, 1= left cheek_____ 2= right cheek____ 3= both_____
   - 5.3. Ear 1= yes _____ 2= no_____
   - 5.4. If yes, 1= left ear_____ 2= right ear_____ 3= both_____
   - 5.5. Lip 1= yes_____ 2= no_____
   - 5.6. If Yes, 1= lower lip_____ 2= upper lip__ 3= both_____
   - 5.7. Leg 1= yes _____ 2= no_____
   - 5.8. If yes, 1= lower limp_____ 2= upper limp_____3= both_____
   - 5.9. Hand 1= yes_____ 2= no_____
   - 5.10. If yes, 1= lower limp_____2= upper limp_____ 3= both_____
   - 5.11. Chin 1= yes_____ 2= no_____
   - 5.12. Nose 1= yes_____ 2= no_____
   - 5.13. Neck 1= yes_____ 2= no_____
   - 5.14. Forehead 1= yes_____ 2= no_____
   - 5.15. Other (specify) _____
6. Suspected leishmaniasis 1= CL_____ 2= MCL_____ 3= DCL_____
7. Scars suggestive of healed leishmaniasis 1= present_____ 2= absent____
8. If cutaneous scar/s present, the location is
   - 8.1. Cheek 1= yes_____ 2= no_____
   - 8.2. If yes, 1= left cheek_____ 2= right cheek____3= both_____
   - 8.3. Ear 1= yes _____ 2= no_____
   - 8.4. If yes, 1= left ear_____ 2= right ear_____ 3= both_____
   - 8.5. Lip 1= yes_____ 2= no_____
   - 8.6. If Yes, 1= lower lip_____ 2= upper lip_____ 3= both_____
   - 8.7. Leg 1= yes _____ 2= no_____
   - 8.8. If yes, 1= lower limp_____ 2= upper limp_____3= both_____

8.9. Hand 1= yes_____ 2= no_____
8.10. If yes, 1= left _____ 2= right _____3= both_____
8.11. Chin 1= yes_____ 2= no_____
8.12. Nose 1= yes_____ 2= no_____
8.13. Neck 1= yes_____ 2= no_____
8.14. Forehead 1= yes_____ 2= no_____
8.15. Other (specify) _____

9. Have you been treated for CL? 1= yes_____ 2=No_____
10. If yes, where were you treated?
1= Traditional healer's house_____ 2= At health center/hospital_____
11. If Q. 10 is yes, how much does the treatment cost in birr per person_____
12. Have you faced any lesion that has self-healed? 1= yes_____ 2=No_____
13. If Q.12 is yes, was the lesion painful/ itchy? 1= yes_____ 2=No_____
12. If Q.12 is yes, was the cause for the lesion physical damage?
1= Yes__ 2=No___
14. Nature of lesion
    1. Ulcer: _____
    2. Nodules: _____
    3. Nodular ulcerated: _____
15. Type of lesion
    1. Wet lesion: _____
    2. Dry lesion: _____
16. Were you bitten by an insect other than a mosquito?
    1. Yes: _____
    2. No: _____
    3. Do not know: _____

If yes, describe the insect _____

17. Signs and symptoms: (circle any that apply)
    1. Pruritus: ____
    2. Pain: _____
    3. Bleeding: _____
    4. Exudation/drainage: _____
    5. Other_____
18. Travel history in the last 6 months
    1. Yes_____
    2. No_____
19. If Q. 18 is yes, period of onset of lesion is
    1. Before travel
    2. After travel
20. If lesion on set is after return, this happened after what period following travel?
    1. less than 1 month: _____
    2. 1- 3 months: _____
    3. 4-6 months: _____
    4. after 6 months: _____

Annexure F: Ethical approval from UNISA

UNISA university of south africa

UNIVERSITY OF SOUTH AFRICA
Health Studies Higher Degrees Committee
College of Human Sciences
ETHICAL CLEARANCE CERTIFICATE

REC-012714-039

HSHDC/457/2015

Date: 25 November 2015 Student No: 5764-754-2

Project Title: Epidemiological investigation of cutaneous Leishmanaisis in Amhara and Southern Nationalities people state, Ethiopia.

Researcher: Lina Gazu Mego

Degree: D Litt et Phil Code: DPCHS04

Supervisor: Prof ZZ Nkosi
Qualification: PhD
Joint Supervisor: Dr S Sibanda

DECISION OF COMMITTEE

Approved [√] **Conditionally Approved** [ ]

Prof L Roets
CHAIRPERSON: HEALTH STUDIES HIGHER DEGREES COMMITTEE

Prof MM Moleki
ACADEMIC CHAIRPERSON: DEPARTMENT OF HEALTH STUDIES

PLEASE QUOTE THE PROJECT NUMBER IN ALL ENQUIRES

Annexure G: Support letter from UNISA, Addis Ababa center

UNISA university of south africa

Date

No

Souther Nation Nationalities People Region Health Bureau

Hawassa

Dear Sir/ Madam,

The University of South Africa(UNISA) extends warm greetings. By this letter , we want to certify that Ms. Lina Gazu Mego(student number 57647542) is a PhD student in the Department of Health Studies at UNISA. Currently, she is at the stage of data collection on her doctoral thesis entitled " Epidemiological investigation of Cutaneous Leishmanaisis in Sodo and Silti Weredas of SNNP, Ethiopia."

This is therfore to kindly request your coopretion in providing the student access to data sources. We would like to thank you in advance for all the assistance that you would provide to her.

Sincerely,

Tsige GebreMeskel Aberra

Deputy Director- Acadamic and ICT Support

University of South Africa
Regional Learning Center
P.OPBox 13836, Addis Ababa, Ethiopia
Telephone: +251 11 435 2244/ +251 11 435 0078
Facsimile: +251 11 435 1242/43/44
Mobile: +251 912 19 1483
www.unisa.ac.za

Annexure H: Collaboration Letter to regional health bureau to conduct research at Sodo district

Date:

To: Southern Nation's Nationalities and Peoples regional health bureau
Hawassa, Ethiopia

Subject: **Support letter**

My name is Lina Gazu Mego, a PhD student in the University of South Africa. I designed a research project entilteld" Epidemiological investigation of Cutaneous Leishmanaisis in Sodo District, SNNP Ethiopia" which will focus on interviewing the community to access knowledge of the disease and identify risk factors for disease transmission. Another componenet of the study deals with clinical sample collction from suspected CL cases for parasite species identification. The findings of this study will provide information on Prevalence of Cutaneous Leishmaniasis in the Districts, Knowledge, attitude, and practices of the community regarding Cutaneous Leishmaniasis, risk factors contribution for the transmission of the disease and Identification of leishmania species circulating in the community. These data will help develop feasible strategy to control of Cutaneous leishmaniasis.

Therefore, I kindly request your kind office to grant me permission to conduct this research at the above-mentioned district.

Lina Gazu Mego (Student number: 57647542)
57647542@mylife.unisa.ac.za

Signature **Date**

Annexure I: Support from the study area

በደ/ብ/ብ/ሕ/ክ/መ በጉራጌ ዞን የሶዶ ወረዳ ጤና ጥበቃ ጽ/ቤት
SNNPRGURAGE ZONE SODO WOREDA
HEALTH OFFICE

ቁጥር / Ref. No. ሶወ 40/6/2014
ቀን/ Date 16-8-09

ጉዳዩ፡ ትብብር እንዲደረግላችዉ ስለመጠየቅ

ከላይ ለመጥቀስ እንደተሞከረዉ ዶ/ር ሲና ጋዙ ዩኒቨርስቲ ኦፍ ሳዉዝ ስፍራካ የፒ.ሄች.ዲ ተማሪ ሲሆኑ በሶዶ ወረዳ ለሚያደርጉት ምርምር ድጋፍ እንዲደርግላችዉ ከክልል በተላከ ደብዳቤ ተጠይቀናል፡፡

በመሆኑም ተመራማሪዋ በምታደርገዉ ጥናት ላይ ስለፈላጊዉ ትብብር እንዲደረግላት በትህትና እንጠይቃለን፡፡

እናቶች በጤና ተቋማት ሲወልዱ
እናት ደህና ልጅም ጤና!!
የመ/ተ/ህ/አገ/ቀ/የስራ ሂደት አስተባባሪ

ግልባጭ

- ለዶ/ር ሲና ጋዙ

Annexure J: Information sheet, consent, parental permission, and assent forms

Information Sheet

Epidemiological investigation of Cutaneous Leishmaniasis in Sodo District of SNNP region, South Ethiopia.

Name of Principal investigator: Lina Gazu Mego
Supervisor: Professor Nikosi zz (University of South Africa)
Co- supervisor: Dr Nigatu Kebede (Addis Ababa University)

Introduction
The purpose of this information sheet and consent form is to describe the research or study that you are being requested to participate in. Before agreeing to participate in the study, please read this form carefully and ask any questions you may have. You are welcome to ask questions at any time after enrolling in the study.

Purpose of the Research Project
The goal of this study is to evaluate indigenous knowledge, disease transmission risk factors, and prevalence of Cutaneous Leishmaniasis (Chewie). This study will entail participation from the entire community of Sodo.

To reduce the risk of infection in the area, the study will provide strategies for developing Cutaneous Leishmaniasis management and prevention approaches. Because this study is one of the few in the country, it will act as a springboard for future research in the country.

Procedure:
We welcome you to participate in this study in order to examine indigenous knowledge attitudes and practices concerning CL, as well as to investigate the disease's prevalence and risk factors in the area. You must understand and sign the agreement form if you wish to participate. Then you'll be interviewed by data collectors, and skin scraping samples will be taken by professional nurses if clinical signs are discovered. For the data collector, you do not need to provide your identity, and your response will be kept private.

Risk/ Discomfort:
- You may experience some discomfort as a result of participation in this research endeavor, particularly in terms of wasting 30 minutes of your time. We hope you will take part in the study for the benefit of the research findings.
- Skin scarping sample taking procedure may be slightly painful.
- If it is your child who is found positive, you may be asked to sign parental form if you consent on participation of your child in the study.
- Your child may pass through slightly painful procedure while the sample is taken.

Incentives/benefits:
There is no benefit/ incentive to you or your child for giving us information for this study. There is no benefit for allowing sample to be taken from the lesion for you or your child. Only positive individuals will get treatment free of charge from ALERT hospital (Addis Ababa) if willing to cover their transport.

Confidentiality:
We will keep the information we collect through this study endeavor private. In order for the completed questionnaire to be anonymous, you are not needed to give your name to the data collectors throughout the interviewing process. The information will be kept in a file that will be locked by the primary Investigator and will not be accessible to anyone else.

Right to refuse or withdraw:
It is absolutely up to you whether or not you participate. You don't have to participate if you don't want to. You won't lose any rewards if you don't. You can withdraw from the study at any time, and your rejection will have no effect on your existing or future relationships with the persons or organizations conducting it. If you do not want to participate, simply let the researcher know and you can opt out at any point during the procedure. If you don't want to, you can choose not to answer any or all of the questions. You can also choose to engage only in the questioner survey and not in the clinical sample collection, or the other way around. You have the option of choosing which study (interview or clinical sampling) your child will take part in. Participation in any of the research is entirely dependent on your and your child's willingness.

Persons to contact:
If you have any question, please contact the following person.

1. Lina Gazu
   Tel: +251-911-07-87-10

If you have read this document or if someone has read this for you and explained to you and if you have been given the chance to ask any questions now or at a later, please sign below.

_____ _____
Name and Signature Date

_____ _____
Signature of a person obtaining consent Date

Consent form

Your signature here confirms that you have read or listened to the study information and that you understand it. Please read the following carefully before signing:

- Purpose of the study
- Study procedure, including
  - ✓ Interview
  - ✓ Sample taking if found to show clinical symptoms (you or your child).

Confidentiality and privacy concerns

The information that we collect from this research project will be kept confidential. You are not required to mention your name for the data collectors in the interviewing process or in the sample taking process so that the filed questionnaire will be anonymous. The information will be stored in a file and locked by the principal Investigator and no other people will have access to it.

By signing, you are agreeing to take part in this research. Put your signature on A if you accept to participate in the interview alone. Please sign B if you agree to participate in the sample collecting procedure. and kindly sign on C if you are willing to participate in both investigations (interview and clinical sample collection). You have the right to withdraw or terminate your participation in the study at any time. Do you agree with me?

I have read and/or listened to the study description and am aware of the procedures and what will happen to me or my child if I participate in the study. I agree to be a part of it. I am aware that I have the option to leave at any moment.

A. I consent to participate in the interview only but not willing sample to be taken from me

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

B. I consent to sample taking from lesion Only, but not willing to be interviewed

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

C. I consent to participate in the interview and sample to be taken from me.

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

Parental Consent Form

Your signature here confirms that you have read or listened to the study information and that you understand it. Please read the following carefully before signing:

- Purpose of the study
- Study procedure, including
  - Clinical examination
  - skin sample
  - biopsy sample
- Risks and benefits of participating in the study
- Right to refuse or withdraw from participation at any time
- Confidentiality and privacy concerns
- Who to contact if you have questions

"By signing, you are agreeing to participate in the study with your child (son/daughter/child, infant/adolescent youth)." You may ask your child to leave the study at any time. Do you agree with me?

I've read and/or listened to the study's explanation and am aware of the procedures and what will happen to my child during the study. I agree to be a part of it. I'm aware that my child has the ability to leave at any time."

If you agree your child participate in interview only, please put your signature on **A**. If you are willing that your child participate in sample collection procedure please sign **B**. and if you are willing that your child participate in both studies (Interview and clinical sample collection), please sign on **C**.

Printed name of (son, daughter/child/infant/adolescent youth) ----------

A. I consent may child to participate in the interview only but not willing sample to be taken from my child

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

B. I consent to sample to be taken from my child Only, but not willing to be interviewed

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

C. I consent my child to participate in the interview and sample to be taken from my child.

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

Assent Form for children between 12 and 17 years of age

I have read and/or listened to the study description and am familiar with the procedures and what will happen to me throughout the study.

My parent(s)/guardian(s) have given me permission to participate in the study, and I agree to do so. I'm aware that I have the option to stop studying at any time.

If you agree to participate in interview only, please put your signature on **A**. If you are willing to participate in sample collection procedure please sign **B**. and if you are willing to participate in both studies (Interview and clinical sample collection), please sign on **C**

A. I consent to participate in the interview only but not willing sample to be taken from me

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

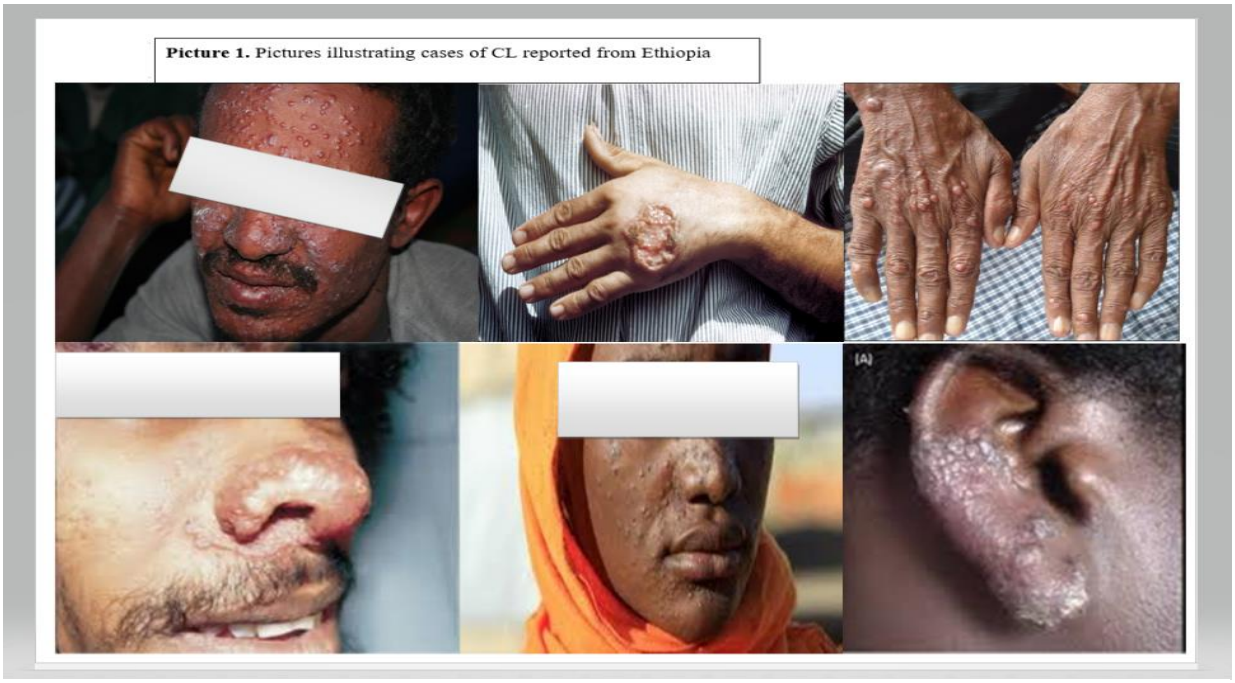
B. I consent to sample taking from lesion Only, but not willing to be interviewed

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

C. I consent to participate in the interview and sample to be taken from me.

Name ----------
Signature---------- Date----------
Signature of investigator/representative---------- Date----------

Annexure K: Picture of CL case and sandfly vector from SNNP, Ethiopia.



Annexure L: Proof reading and editing confirmation letter

EDITING AND PROOFREADING CERTIFICATE

7542 Galangal Street

Lotus Gardens

Pretoria

0008

17 November 2020

TO WHOM IT MAY CONCERN

This certificate serves to confirm that I have edited Lina Gazu Mego's thesis entitled, **EPIDEMIOLOGICAL INVESTIGATION OF CUTANEOUS LEISHMANIASIS IN SODO DISTRICT SOUTHERN ETHIOPIA.**

I found the work easy and intriguing to read. Much of my editing basically dealt with obstructionist technical aspects of language, which could have otherwise compromised smooth reading as well as the sense of the information being conveyed. I hope that the work will be found to be of an acceptable standard. I am a member of Professional Editors' Guild.

Hereunder are my particulars:

Jack Chokwe (Mr)

Contact numbers: 072 214 5489

jackchokwe@gmail.com

Professional EDITORS Guild

Annexure M: Turniting plagiarism report

Document Viewer

Turnitin Originality Report

Processed on: 03-Nov-2021 18:48 SAST
ID: 1692088550
Word Count: 76449
Submitted: 1

EPIDEMIOLOGICAL INVESTIGATION OF CUTANEOUS LE... By Lina Gazu Mego

Similarity Index	Similarity by Source	
21%	Internet Sources:	19%
	Publications:	13%
	Student Papers:	9%

exclude quoted | exclude bibliography | exclude small matches | mode: quickview (classic) report | Change mode | print | download

2% match (Internet from 27-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 26-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 27-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 27-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 12-Oct-2021)
http://etd.aau.edu.et

<1% match (Internet from 26-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 11-Oct-2021)
http://etd.aau.edu.et

<1% match (Internet from 23-Aug-2020)
http://etd.aau.edu.et

<1% match (Internet from 11-Oct-2021)
http://etd.aau.edu.et

<1% match (Internet from 12-Oct-2021)
http://etd.aau.edu.et

<1% match (Internet from 27-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 11-Oct-2021)
http://etd.aau.edu.et

<1% match (Internet from 27-Sep-2021)
http://etd.aau.edu.et

<1% match (Internet from 27-Sep-2021)

“In God we trust, all others must bring data. “
W.E Deming