Woody Plant Species Diversity, Land Cover Dynamics and Ethnobotany of Medicinal Plants in Habru District, Amhara Region, Ethiopia

By

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GRADUATE PROGRAMMES

This is to certify that the dissertation prepared by Mulugeta Alemu Molla, entitled: **Woody Plant Species Diversity, Land Cover Dynamics and Ethnobotany of Medicinal Plants in Habru District, Amhara Region, Ethiopia.** And submitted in fulfillment of the requirements for the Degree of Doctor of Philosophy in Plant Biology and Biodiversity Management complies with the regulations of the University and meets the accepted standards with respect to originality and quality.

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Dedication

This research is dedicated to my family, my wife **Muluemebet Semaw**, and our daughter, **Mariham Mulugeta**. Their constant support and encouragement have been pivotal in my academic pursuits. Their enduring love and faith in my abilities have been instrumental in overcoming obstacles and achieving success.

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List of Acronyms/Abbreviations

AAU	Addis Ababa University
AHRI	Armauer Hansen Research Institute
ArcGIS	Aeronautical Reconnaissance Coverage Geographic Information System
BA	Basal Area
CBD	Convention on Biological Diversity
CSA	Central Statistical Agency of Ethiopia
DBH	Diameter at Breast Height
DMR	Direct Matrix Ranking
DPBBM	Department of Plant Biology and Biodiversity Management
EBI	Ethiopian Biodiversity Institute
EFAP	Ethiopian Forestry Action Program
ENMA	Ethiopian National Meteorology Agency
ESRI	Environmental Systems Research Institute
ETH	Ethiopian National Herbarium
ETM+	Enhanced Thematic Mapper
FAO	Food and Agricultural Organization of the United Nations
FEE	Flora of Ethiopia and Eritrea
FGD	Focus Group Discussion
FL	Fidelity Level
GCP	Ground Control Points
GDARDO	Gubalafto District. Agriculture and Rural Development Office
GIS	Geographic Information System
GPS	Geographic Positioning System
ha	Hectare
HDARDO	Habru District Agriculture and Rural Development Office
HHs	Households
IBC	Institute of Biodiversity Conservation
ICF	Informant Consensus Factor
IUCN	The International Union for Conservation of Nature

IVI	Important Value Index
km	Kilo meter
LC	Land Cover
LU	Land Use
LULC	Land Use Land Cover
m.a.s.l	Meter Above Sea Level
MAPA	Methods of Preparation and Application
MPs	Medicinal Plants
NDVI	Normalized Difference Vegetation Index
OLI	Operational Land Imager
PR	Preference Ranking
RD	Relative Density
RDo	Relative Dominance
RF	Relative Frequency
RV	Riverine Vegetation
SAC	Species Accumulation Curve
TM	Traditional Medicine
USGS	United States Geological Survey
UTM	Universal Transverse Mercator
UV	Use Value
WHO	World Health Organization

Summary

Geospatial techniques are vital for examining the dynamics of land use land cover (LULC) in specific areas. In Ethiopia, a blend of human activities and natural factors is threatening the vegetation, leading to the degradation and fragmentation of forests. The people of Habru District in the North Wollo Zone of Amhara Region, Ethiopia, have a rich tradition of utilizing local plant resources for a variety of needs, particularly for traditional herbal remedy preparation and use. However, the district has not been adequately studied for its land use dynamics, ecological and ethnobotanical resources linked with the associated knowledge. This gap highlights the necessity for detailed studies on woody plant species diversity in relation to land use land cover dynamics and ethnobotanical study of medicinal plants and their uses within the study area. Moreover, Ethiopia's culture on the utilization of traditional medicine as a primary healthcare system for treating human diseases calls for empirical research to validate these practices against infectious diseases. Therefore, this study was aimed to examine the LULC change dynamics in Habru District for a period of 36 years (1985 – 2021), assessing the woody species composition, structure and plant community analysis of Gerado, Micha and Mekelet Forest Patches, documenting and analyzing medicinal plant species used by the local communities to treat human ailments along with the associated traditional knowledge and practices and empirically validating the efficacy of selected traditional herbal remedies used by the local people at present.

This study, employed remote sensing methods to analyze the temporal and spatial changes in land use land cover within the study area, using Landsat images from 1985 - 2021. To complement this, Focus Group Discussions (FGDs) and interviews with key informants were carried out to collect insights on the factors influencing LULC changes. Image classification was performed using the supervised classification technique, specifically employing the minimum likelihood algorithm for land use mapping. For vegetation analysis, a major category consisting of all the woody species (trees and shrubs) having $\geq$ 2.5 cm diameter at breast height were collected from a total of 95 plots (20 x 20 m^2) laid systematically from Gerado (43 plots), Micha (28 plots) and Mekelet (24 plots) forest patches. The Shannon-Wiener diversity index was used to quantify the woody species richness and evenness of the forest patches. This study also determined plant community types and examined their correlation with environmental factors using agglomerative hierarchical cluster analysis and Redundancy Analysis (RDA) in the R

software environment. The ethnobotanical data were collected using semi-structured interviews, guided field walks, focus group discussions, and market surveys. A total of 388 informants (250 males and 138 females) were selected from all 13 kebeles (subdistricts) within Habru District using systematic random sampling technique, and 42 key informants were purposively selected in each from among the randomly identified informants. Descriptive statistics, preference ranking, direct matrix ranking, informant consensus factor and fidelity level were applied for data analysis. For antimicrobial and phytochemical screening, the candidate medicinal plants were selected considering their high informant consensus and fidelity level values for their efficacy in treating infectious diseases in the area. Ethanol and aqueous extracts were prepared from the plant materials, and their antibacterial activities were evaluated against standard gram-positive and gram-negative bacterial strains. The minimum inhibitory concentration (MIC) was determined using the broth dilution method. Furthermore, phytochemical screening was performed using standard qualitative tests to identify various secondary metabolites.

The study results revealed that there was a substantial expansion of settlement areas in Habru District, with a notable decrease in shrubland and forestland. Conversely, cultivated land and settlement areas increased, indicating significant anthropogenic impacts on natural landscapes. This study investigated the primary drivers of LULC change in Habru District. The analysis identified cultivated land expansion, settlement area expansion, charcoal production, firewood collection, and tree cutting as the major proximate factors influencing LULC change. Additionally, the study explored underlying causes, including population growth/pressure, land tenure insecurity, economic challenges (poverty and unemployment), and climate change (precipitation variability). A total of 55 woody plant species were recorded from the three forest patches. The study also identified four distinct plant community types, influenced by factors like altitude and human activities such as tree cutting. Of these, 35 woody plant species, constituting (64%), were identified as medicinal plants used by the local people. Furthermore, a total of 134 medicinal plant species in 110 genera and 54 families used in traditional medicine against human ailments in Habru District were documented, highlighting the district's significance in biodiversity conservation and primary healthcare delivery. High consensus factors for gastrointestinal and parasitic ailments (ICF = 0.85), febrile diseases (ICF = 0.84), and cultural conditions (ICF = 0.84), highlight the community's reliance on medicinal plants. Multipurpose plants such as *Solanum somalense* Franchet. (91.3% FL), *Ocimum lamiifolium* Hochst. ex. Benth.

(88.9% FL), and *Verbascum sinaiticum* Benth. (85.7% FL) exhibited notable healing potentials. Of the documented medicinal plants, only 16.4% are marketable, including species like *Terminalia brownii* Fresen. *Myrtus communis* L., *Ruta chalepensis* L., *Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif., *Allium sativum* L. and *Capsicum annuum* L., commonly found in local markets. Ethanol extracts of selected plants (*Solanum somalense* Franchet., *Withania somnifera* (L.) Dunal and *Calpurnia aurea* (Ait.) Benth.) showed promising antibacterial activity against specific bacterial strains. The presence of secondary metabolites like steroids, alkaloids, and flavonoids underscores the pharmacological potential of these plants.

The study underscores the profound impact of anthropogenic activities on natural ecosystems and the crucial role of traditional knowledge in leveraging biodiversity for healthcare. The study emphasizes the critical need for strategies addressing LULC change drivers to protect natural resources and ensure sustainable ecosystem services to contribute to livelihoods. It highlights the inadequacy of current management in conserving forest patches and suggests tailored, community-based conservation to improve ecosystem resilience. Recognizing the deep connection between communities and medicinal plants, the study advocates for biodiversity conservation, sustainable healthcare, and the preservation of traditional knowledge, underscoring the mutual dependence of human societies and natural environments.

CHAPTER ONE

1. General Introduction

1.1. Background of the study

Biodiversity encompasses the wide array of life forms that inhabit our planet, including plants, animals, fungi, microorganisms, Monera (bacteria), and Protista (single-celled organisms) (Purvis and Hector, 2000). Biodiversity plays a crucial role in maintaining the stability and resilience of ecosystems, underpinning ecosystem services that are vital for human well-being, including food security, disease regulation, and climate stability. Recent studies indicate the importance of biodiversity in enhancing ecosystem productivity and stability, as well as its contribution to carbon sequestration and storage, which are pivotal in mitigating climate change impacts (Díaz *et al.*, 2019). Furthermore, biodiversity offers tremendous economic, ethical, and aesthetic advantages, underpinning ecosystem health and providing the genetic basis for essential crops and medicines (Tilman, 2000). The preservation of biodiversity is not only a conservation imperative but also a critical component in sustainable development strategies aimed at maintaining ecological balance and ensuring a sustainable future for all life forms on Earth.

Land use and land cover (LULC) change represents a fundamental component in understanding the dynamics of Earth's surface and its impact on global environmental changes, including biodiversity loss and climate change (Lambin *et al.*, 2003). The interplay between human activities and natural ecosystems has led to significant alterations in the Earth's vegetation cover, a critical aspect of vegetation ecology that influences both global biodiversity and human well-being (Foley *et al.*, 2005). In the context of Africa, these changes are particularly pronounced due to a combination of factors including population growth, agricultural expansion, and deforestation, which collectively threaten the continent's rich biodiversity and ecological integrity (Brink and Eva, 2009). Changes in land use and cover, particularly the conversion of natural vegetation into farmlands, grazing lands, infrastructure, and human settlements, play a substantial role in the loss of biodiversity and the disruption of ecosystem services.

Vegetation is a fundamental aspect of biodiversity, serves as a critical physical manifestation of ecosystems, forming the foundation of ecological pyramids and providing habitats for other elements within biodiversity (Kent, 2011). The Ethiopian highlands predominantly consist of

Dry Afromontane forests, making up the most extensive area of such vegetation (Lillesø *et al.*, 2011). The country's rich biodiversity is a result of its varied landscapes, climate zones, and ecosystems, making it one of Africa's most species-diverse nations with over 5,757 vascular plant species, with approximately 10% endemic to the flora area (Sebsebe Demissew *et al.*, 2021). This diversity includes a vast array of plant species that hold significant ecological, economic, and medicinal importance (Ensermu Kelbessa and Sebsebe Demissew, 2014). This biodiversity is under threat from anthropogenic activities, climate change and unsustainable land use practices (EBI, 2022). Moreover, disturbances caused by human activities have been identified as significant factors leading to the degradation of both the structure and the floral diversity of these forests (Simachew Bantigegn, 2020; Woods *et al*., 2020; Tayachew Birhanu *et al.*, 2021).

The utilization of medicinal plants, deeply embedded in Ethiopian culture, underscores the importance of ethnobotanical studies in preserving traditional knowledge and biodiversity (Ashenafi Osman *et al.*, 2020). The review by Nigatu Tuasha *et al.* (2018) showed that about 80% of Ethiopia's population relies on traditional medicine, primarily derived from plants, for healthcare needs. This reliance not only highlights the cultural significance of medicinal plants but also underscores the urgency of documenting and conserving Ethiopia's botanical resources amidst ongoing environmental changes.

The outcomes of phytochemical analyses and tests for antimicrobial efficacy of medicinal plants in Ethiopia serve multiple purposes in traditional medicine. These include confirming the historical applications of these plants, discovering new potentially active compounds, enhancing product formulations, reinforcing cultural traditions, and combating antimicrobial resistance (Sileshi Dubale *et al.*, 2023). Furthermore, given the global rise in antibiotic resistance, the exploration of alternative sources of antimicrobial agents from plants offers promising avenues for new drug development (Keita *et al.*, 2022). Medicinal plants are a rich source of bioactive compounds, including alkaloids, flavonoids, terpenoids, and phenolic compounds, which exhibit diverse biological activities, including potent antimicrobial properties (Keita *et al.*, 2022; Abdallah *et al.*, 2023). Ethiopia's rich flora, with its vast array of medicinal plants, provides a valuable resource for such investigations, contributing to global efforts in discovering novel therapeutic compounds (Dilbato *et al.*, 2019; Admasu Moges and Yohannes Moges, 2020).

Like elsewhere in Ethiopia, the people of Habru District in the North Wollo Zone of the Amhara Regional State exemplifies the intricate relationship between humans and their environment. This district, characterized by its diverse ecological settings and traditional agricultural practices, serves as an ideal study area for examining the interconnections between land use changes, vegetation ecology, ethnobotanical knowledge, and the antimicrobial properties of biologically active medicinal plants. Despite its rich biodiversity and cultural heritage, the district faces challenges related to anthropogenic factors, environmental degradation, and the sustainability of its natural resources. This study aims to bridge these gaps by conducting a comprehensive analysis of the district's floristic composition, land use practices, and the ethnobotanical knowledge of its inhabitants, thereby contributing valuable insights to the fields of conservation biology, sustainable development, and ethnopharmacology.

1.2. Statement of the problem

Forest ecosystems fulfill several vital functions within the environment through the provision of a various array of goods, medicine, and ecological advantages (Mori *et al.*, 2017). They provide habitats for a wide variety of flora and fauna, which depend on these wooded regions for their existence (Brockerhoff *et al.*, 2017). Moreover, forests function as carbon reservoirs, capturing and sequestering atmospheric carbon dioxide, thereby playing a significant role in counteracting climate change through the reduction of carbon emissions (Perry *et al.*, 2008). In many developing countries like Ethiopia, most of the rural populations directly live on land resource as a source of livelihood (Babulo *et al.*, 2009). In Ethiopia, forests play a key role in supporting both local livelihoods and the national economy by supplying resources such as timber, fuel wood, fodder, and various minor products. Additionally, they bolster the agricultural sector by enhancing soil fertility and preventing erosion (Demel Teketay *et al.*, 2010; Melkie Achenef, 2020). The FAO report on forest resources in Ethiopia highlights the country's diverse vegetation resources, which range from lowland scrubs to tropical rainforest (FAO, 2016). Ethiopia's forest resources include both natural and planted forests, with natural forests consisting of moist and dry tropical Afromontane forests, woodlands, and shrub lands (Demel Teketay *et al.*, 2010; EBI, 2022). Even though diverse ecosystems of Ethiopia have endowed the country with a various biological wealth of plants and animals, for a variety of reasons, these resources are under severe threats. According to the FAO report the country's natural forest is declining and characterized by high rate of forest degradation. A century ago, around 1900, approximately 40% of Ethiopia's land was covered by forests. By 1975, forest cover had drastically reduced to about 4% (FAO, 2010). Recent reports, however, indicate that Ethiopia's forest cover has increased to 15% due to reforestation and conservation efforts (EBI, 2022). The major reason behind degradation of forests is human interference such as expansion of agricultural land, grazing, firewood collection and poverty (Yohannes Kidane *et al.*, 2012; Alemayehu Urgesa *et al.*, 2016; EBI, 2022). Furthermore, population growth is one of the reasons for the sever problems of environmental destruction and land use change, that the country is facing today (Agumassie Genet, 2020). Rapid population growth can create pressure on basic resources such as water, forests, and land, as well as biodiversity beyond what is available (Mathewos Muke, 2019). Moreover, human activities including agricultural expansion, grazing, and charcoal production and firewood collection, pose significant threats to Ethiopia's remaining natural forests (Tamrat Bekele, 1993; Edwards *et al.*, 1999; Tsehaye Gebrelibanos and Mohammed Assen, 2015). It is suggested that integrating spatiotemporal studies with ecological and ethnobotanical data is crucial for effective conservation and development strategies. This comprehensive approach ensures a well-rounded understanding of a given area's needs and potentials for sustainable management (Martin, 1995; Araújo and Ferraz, 2014). They necessitate prompt and precise scientific actions to ensure the successful preservation and restoration of natural forest ecosystems.

In the Habru District of Ethiopia, various interrelated challenges threaten the ecological integrity of the district. These challenges encompass a spectrum of issues, including rapid land use and land cover changes, biodiversity loss, and the depletion of valuable plant resources essential for both local livelihoods and healthcare delivery. Habru District, like many regions in Ethiopia, is experiencing a decline in forest cover due to land use and land cover changes driven by factors like agricultural expansion, grazing, and firewood collection (Yohannes Kidane *et al.*, 2012; Alemayehu Urgesa *et al.*, 2016; EBI, 2022). This loss of biodiversity coincides with a potential erosion of indigenous knowledge about medicinal plants and their uses. Local communities in Habru District rely heavily on these plants for their primary healthcare needs due to limited access to conventional medicine. This traditional knowledge system is often passed down through generations, but with societal changes and population growth, this valuable information may be disappearing (Agumassie Genet, 2020). Furthermore, the ongoing loss of indigenous and local knowledge regarding medicinal plants exacerbates these problems, highlighting a critical need for conservation efforts and documentation of traditional ethnobotanical knowledge. Moreover, the reliance of local communities on plant-based healthcare further indicates the significance of preserving both the ecological and cultural dimensions of the district's flora. This study aims to investigate the patterns and dynamics of land use and land cover changes over a 36-year period (1985–2021), alongside investigating the composition, structure, and ecological significance of forest patches. Additionally, it seeks to document the ethnobotanical knowledge associated with medicinal plants, contributing valuable insights into conservation biology, sustainable development, and ethnopharmacology through antimicrobial and phytochemical screening of selected medicinal plants. By recognizing the intrinsic value of indigenous knowledge systems and the multifaceted role of plant resources, this research endeavors to inform evidence-based conservation strategies and promote the sustainable utilization of natural resources in the Habru District, North Wollo, Amhara Region, Ethiopia.

1.3. Research questions and hypotheses

1.3.1. Research questions

1. What does spatiotemporal land use land cover change of Habru District look like from 1985 to 2021, and what are the major driving forces for the change?
2. What are the floristic composition, community type and diversity of plant species existing in the forest patches of Habru District and its environs located in North Wollo Zone, Amhara Region, Ethiopia?
3. What are the traditional medicinal plants used by indigenous people living in Habru Districts? Which of these plants are used to treat human ailments in the area?
4. Do traditional medicinal plants with high informants' consensus and fidelity level values show positive results when subjected to activity tests against microbial agents causing infectious diseases?
5. What are the main threats to medicinal plants and associated indigenous and local knowledge in the area? Are there local conservation practices?

1.3.2. Research hypotheses

1. A significant portion of the natural forests of the study area has been lost due to LULC change and the major drivers are population growth and settlement; agricultural expansion; deforestation and land clearing; and firewood collection.
2. The natural forest patches found in the fragmented landscapes of Habru District and its environs have high woody species diversity and serve as reservoirs of useful plants.
3. People in the study area are knowledgeable on use and management of traditional medicinal plants to treat human ailments.
4. Traditional medicinal plants with highest informants' consensus and fidelity levels on their local uses against microbial-based infectious diseases are best candidates for possible use as antimicrobial agents.
5. The people in Habru District have traditional management practices and cultures that help to ward off the threats and contribute to the management and conservation of medicinal plants diversity and/or associated knowledge in the study area.

1.4. Objectives of the study

1.4.1. General objective

- The general objective of this research was to evaluate land use land cover dynamics, document and analyze the current woody plant species diversity and investigate ethnobotanical knowledge associated with the medicinal plants used by the people of Habru District, North Wollo Zone, Amhara region, Ethiopia.

1.4.2. Specific objectives

The research targets to:

1. Analyze temporal and spatial land use land cover dynamics from 1985 to 2021 and identify the major driving forces behind the land cover dynamics in the study area.
2. Examine the status of floristic composition, diversity, structure, and major plant community types of selected forest patches in Habru District of North Wollo Zone, Amhara region Ethiopia.
3. Gather, record and document indigenous and local knowledge of the people on medicinal plants against human ailment in the study area.
4. Investigate antimicrobial property of widely reported medicinal plants based on the degree of informants' consensus and fidelity level values for their use to treat infectious diseases.
5. Analyze the contributing factors to the depletion or conservation of medicinal plants and the associated knowledge in the target study area.

1.5. Conceptual framework

The overall conceptual framework of the study and linkages among the major components and sub-components of the study are diagrammatically shown in Figure 1.

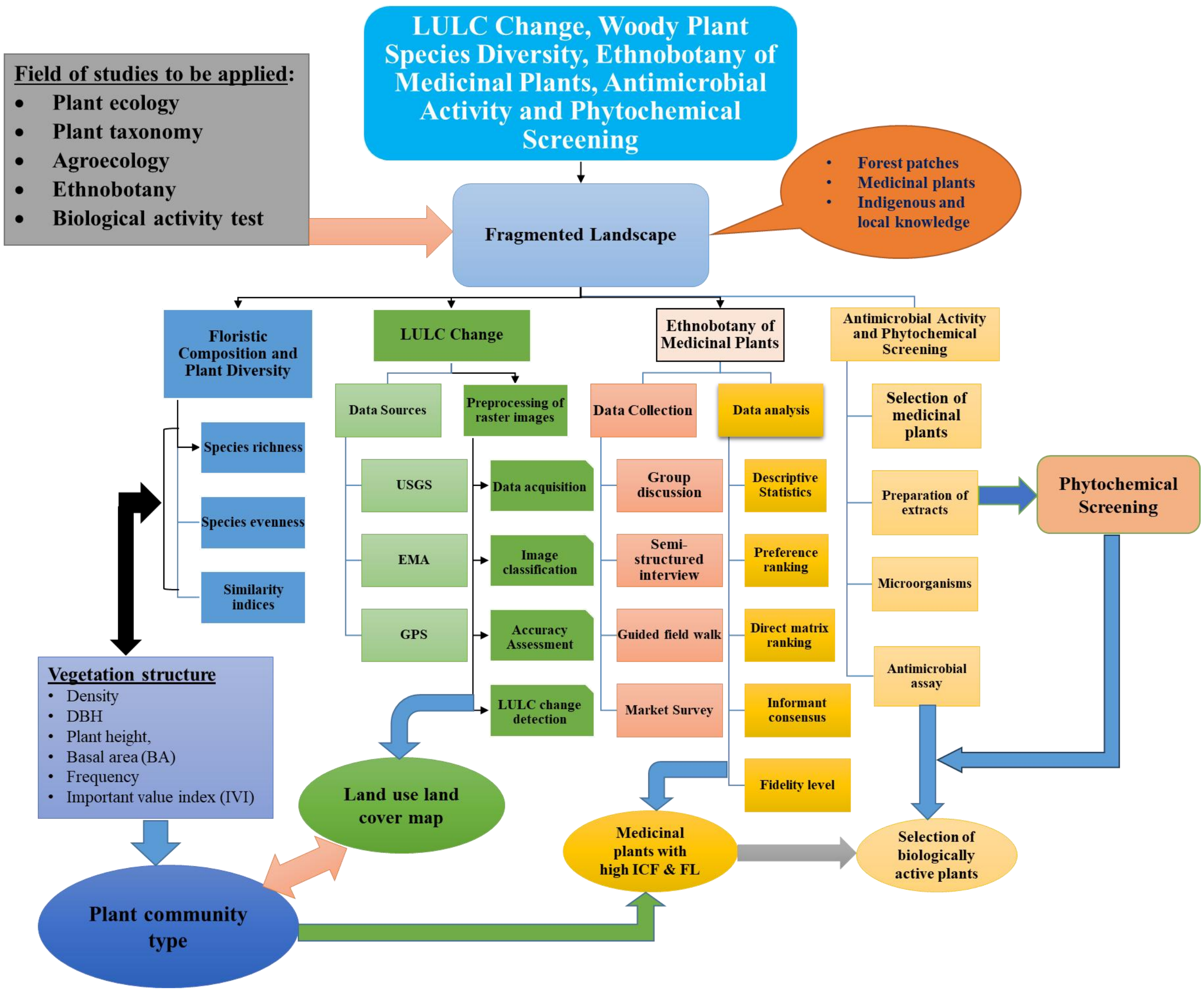

Figure 1: Links among the different components and sub-components of the study

1.6. Structure of the dissertation

This dissertation, structured into six comprehensive chapters, presents an extensive investigation into various ecological and ethnobotanical aspects. Each chapter focuses on a distinct component of the study, contributing collectively to the overarching research objectives focusing on land use land cover change dynamics, woody plant species diversity, ethnobotanical uses of medicinal plants against human ailments and analyzing antimicrobial activities and phytochemical screening of selected medicinal plants used in Habru District.

Chapter 1: Introduction: This chapter is the general introduction which delineates the central research questions, hypotheses, and objectives, underscoring the significance of the study.

Additionally, this chapter outlines the scope and limitations encountered during the research process. A clear presentation of the dissertation's structure is also provided, guiding the reader through the subsequent chapters.

Chapter 2: Land use and land cover change study: The second chapter delves into the first component of the research, analyzing the dynamics of land use and land cover changes. This section employs a combination of remote sensing techniques and field observations to elucidate the patterns and drivers of these changes, offering insights into their ecological and socio-economic implications.

Chapter 3: Woody plant species composition, structure, and plant community type study: The third chapter presents an exhaustive study of floristic composition, structure, and plant community types of forest patches. This component involves detailed field surveys and statistical analyses to understand the distribution, diversity, and ecological characteristics of plant communities within the study area.

Chapter 4: Ethnobotanical study of medicinal plants: In the fourth chapter, the focus shifts to an ethnobotanical study of medicinal plants. This section incorporates local knowledge and practices, documenting the use of medicinal plants by indigenous communities. It explores the cultural significance, traditional uses, and potential therapeutic properties of these plants.

Chapter 5: Antimicrobial and phytochemical analysis study: The fifth chapter discusses the antimicrobial and phytochemical analysis of selected medicinal plants. This component combines laboratory-based phytochemical screenings with antimicrobial tests, aiming to validate the traditional medicinal uses and identify bioactive compounds.

Chapter 6: General discussion, conclusions, and recommendations: The final chapter synthesizes the findings from the previous chapters, offering a comprehensive discussion on the overall implications of the study. It articulates the conclusions drawn from the research and proposes recommendations for future studies and policy interventions. Major articles published from this study are listed below:

1. Mulugeta Alemu, Zemede Asfaw, Ermias Lulekal, Bikila Warkineh, Asfaw Debella, Bihonegn Sisay and Eyob Debebe (2024). Ethnobotanical study of traditional medicinal plants used by the local people in Habru District, North Wollo Zone, Ethiopia. In Journal of Ethnobiology and Ethnomedicine (Vol. 20, Issue 1). Springer Science and Business Media LLC. https://doi.org/10.1186/s13002-023-00644-x (Article published).

2. Mulugeta Alemu, Ermias Lulekal, Zemede Asfaw, Bikila Warkineh, Asfaw Debella, Abiy Abebe, Sileshi Degu and Eyob Debebe (2024). Antibacterial Activity and Phytochemical Screening of Traditional Medicinal Plants Most Preferred for Treating Infectious Diseases in Habru District, North Wollo Zone, Amhara Region, Ethiopia. PLoS ONE 19(3): e0300060. https://doi.org/10.1371/journal.pone.0300060 (Article published).
3. Mulugeta Alemu, Bikila Warkineh, Ermias Lulekal, and Zemede Asfaw (2024). Woody Species Composition, Structure and Plant Community Analysis of Gerado, Micha and Mekelet forest patches in North Wollo Zone, Amhara Region, Ethiopia **(Article under review).**
4. Mulugeta Alemu, Bikila Warkineh, Ermias Lulekal and Zemede Asfaw (2024). Analysis of Land Use Land Cover Change Dynamics in Habru District, Amhara Region, Ethiopia. **(Article under review).**

CHAPTER TWO

2. Analysis of Land Use Land Cover Change Dynamics in Habru District, Amhara Region, Ethiopia

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A paper under review

Analysis of Land Use Land Cover Change Dynamics in Habru District, Amhara Region, Ethiopia

Abstract

This study examines spatiotemporal changes in Land Use Land Cover (LULC) patterns within Habru District, Amhara Region, Ethiopia, between 1985 and 2021. Employing Landsat imagery and a supervised classification approach, we mapped six LULC classes: water body, settlement area, cultivated land, bare land, shrub land, and forest land. Ground Control Points were obtained through field observations to ensure image accuracy. Complementing the remote sensing analysis, focus group discussions (FGDs) and key informant interviews were conducted to understand local perspectives on LULC changes. The analysis revealed significant LULC transformations over the study period. Between 1985 and 2021, there was a significant increase in cultivated land and settlement areas, which expanded from 12.27% and 0.23% to 42.12% and 2.58% of the total area, respectively. Conversely, shrub and forest lands saw a marked reduction during the same period, declining from 71.52% and 12.88% to 45.35% and 7.06% of the total area, respectively. Water body area showed a minor increase, while bare land exhibited negligible change. Agricultural land and settlement area expansion, population growth, land tenure insecurity, economic challenge, and climatic change were identified as key drivers of observed LULC changes. Our findings underscore the urgency of implementing multifaceted interventions to address these drivers and promote sustainable resource use practices. Such actions are critical for safeguarding Habru District's natural resources and ensuring the long-term viability of ecosystem services.

Key Words: Habru, Landsat, LULC, Remote sensing, Spatiotemporal dynamics

2.1. Introduction

Land Use Land Cover (LULC) change is a complex phenomenon influenced by biophysical and socio-economic factors (Arsanjani, 2012). These factors encompass long-term climatic shifts, geomorphological and ecological processes, alterations in vegetation cover and landscape structures, climate variability, and the greenhouse effect (Lambin and Strahler, 1994; Mertens and Lambin, 1999; Rindfuss *et al.*, 2004; Arsanjani, 2012). Land use (LU) refers specifically to the human modification and application of land for economic purposes (Turner *et al.*, 1994; Sohl and Sleeter, 2012). LU reflects the actions taken to shape the land cover and the resulting economic benefits derived. Land cover (LC), in contrast, refers to the continuous natural characteristics of the Earth's surface, such as vegetation, rocks, soil, water and human-made structures (Mölders, 2011).

Land use land cover (LULC) change is a globally recognized environmental concern due to its far-reaching consequences. These consequences include biodiversity loss, increased pollution, and heightened climate variability (Lambin *et al.*, 2006; Mahmood *et al.*, 2010). Furthermore, LULC changes can disrupt crucial ecosystem services by altering moisture and temperature regimes (Mölders, 2011; Wang *et al.,* 2015). Remote sensing, which involves the use of aerial sensors to gather data on Earth's surface, atmosphere, and oceans, plays a vital role in monitoring and analyzing LULC changes (Sohl and Sleeter, 2012). Remote sensing data allows researchers to understand and predict these landscape transformations (Petit and Lambin, 2001). The multifaceted impacts of LULC changes encompass a wide range of issues. These include reduced provisioning services: food, fiber, and timber production (Chhabra *et al.*, 2006), increased disease risks, elevated atmospheric gasses contributing to climate change, diminished biodiversity, life support functions, and agrobiodiversity, soil degradation and altered freshwater hydrology, agricultural water use, and coastal zones (Chhabra *et al.*, 2006).

In Sub-Saharan Africa, rapid population expansion has led to the excessive utilization of natural resources (Lambin, 2001). A complex interplay of biophysical processes and socioeconomic factors shapes LULC patterns across space and time (FAO, 2016). In developing countries like Ethiopia, agriculture, the mainstay of the economy (FAO, 2016), is a key driver of LULC change. Ethiopia's agricultural expansion has significantly altered natural landscapes, including forests and grasslands bordering cultivated and grazing areas (Lambin *et al.*, 2003). However, recent efforts towards conservation and rehabilitation offer a glimmer of hope for reversing these trends.

Forests, a critical natural resource, are particularly vulnerable to LULC changes, often being converted to agricultural land (Lambin, 2001). Several studies conducted across Ethiopia pinpoint population growth, settlement expansion, agricultural intensification, deforestation, land clearing, and fire as the primary drivers of LULC change (Yohannes Kidane *et al.*, 2012; Alemayehu Urgesa *et al.*, 2016). Studies in various regions of the country support this observation, including: northwestern Ethiopia (Gete Zeleke, 2000); Kalu District of southern Wollo (Kebrom Tekle and Hedlund, 2000); northwestern Ethiopia (Gete Zeleke and Hurni, 2001); eastern Tigray (Fikir Alemayehu et al., 2009); Lake Alemaya watershed in eastern Ethiopia (Assen, 2011); Didessa sub-basin (Terefe Tolessa *et al.*, 2020) and the Afroalpine vegetation belt of the Abune Yosef mountain range in the north (Kflay Gebrehiwot, 2020).

Besides population increase, significant challenges to Ethiopia's remaining natural forests arise from human-induced threats such as deforestation for agricultural and grazing land expansion, along with the uncontrolled harvesting of timber and fuel wood (Tamrat Bekele, 1993; Edwards *et al.*, 1999; Tsehaye Gebrelibanos and Mohammed Assen, 2015). This is a major concern for Habru District and other similarly affected areas. The status of LULC changes of Habru District in North Wollo Zone remained undocumented. This research aims to assess and quantify the spatial changes in LULC within the Habru over the past 36 years (1985–2021) and to identify the key drivers of these changes. Understanding these LULC dynamics is critical for informing the development and implementation of sustainable natural resource management strategies in Habru District. Furthermore, the outcomes of this research will provide valuable insights to decision-makers, enabling them to comprehend the extent of changes in the study area and make informed decisions based on these findings.

2.2. Materials and methods

2.2.1. Description of the study area

The study was conducted in Habru District, North Wollo Zone, Amhara Region, Ethiopia (Figure 1.1). The district encompasses 36 rural kebeles (sub-districts) and three urban kebeles, spanning a total area of approximately 1350.4322 square kilometers. Mersa, the district's central town, lies 491 kilometers north of Addis Ababa, the capital of Ethiopia, 406 kilometers east of Bahirdar, the capital of the Amhara Region, 90 kilometers northeast of Dessie, the capital of South Wollo Zone, and is located between latitudes 11°35'0" to 11°55'0"N and longitudes 39°30'10" to

40°1'0"E. The district's elevation ranges from 1430 to 2800 meters above sea level (Zelalem Teshager *et al.*, 2018). Habru is demarcated to the south by the Mille River, which delineate it from the South Wollo Zone to the west by Gubalafto District, to the north by the Alewuha River, marking its boundary with Kobo District, and to the east by the Afar Region (HDARDO, 2019). The climatic conditions in Habru District are diverse, featuring a coldest climate zone known locally as DEGA or cool and humid (2300 – 3200 m.a.s.l), comprising 3.5% of the district's area, a warmest climate zone referred to as KOLLA or warm semi-arid (500 – 1500 m.a.s.l), covering 56.5%, and a medium climate zone named WEINADEGA or cool sub-humid (1500 – 2300 m.a.s.l), constituting 40% of the area. The average annual temperature recorded in the study area is 20.1°C, aligning with observed monthly minimum and maximum temperatures of 10.6°C and 30.8°C, respectively (ENMA, 2019). The climate diagram showed that the study area has a bimodal pattern of rainfall distribution, spanning from March to October, peaking during the spring months of March and April, and the summer period from July to October. The mean annual rainfall is approximately 1045 mm/year, with a dry season lasting from November to February.

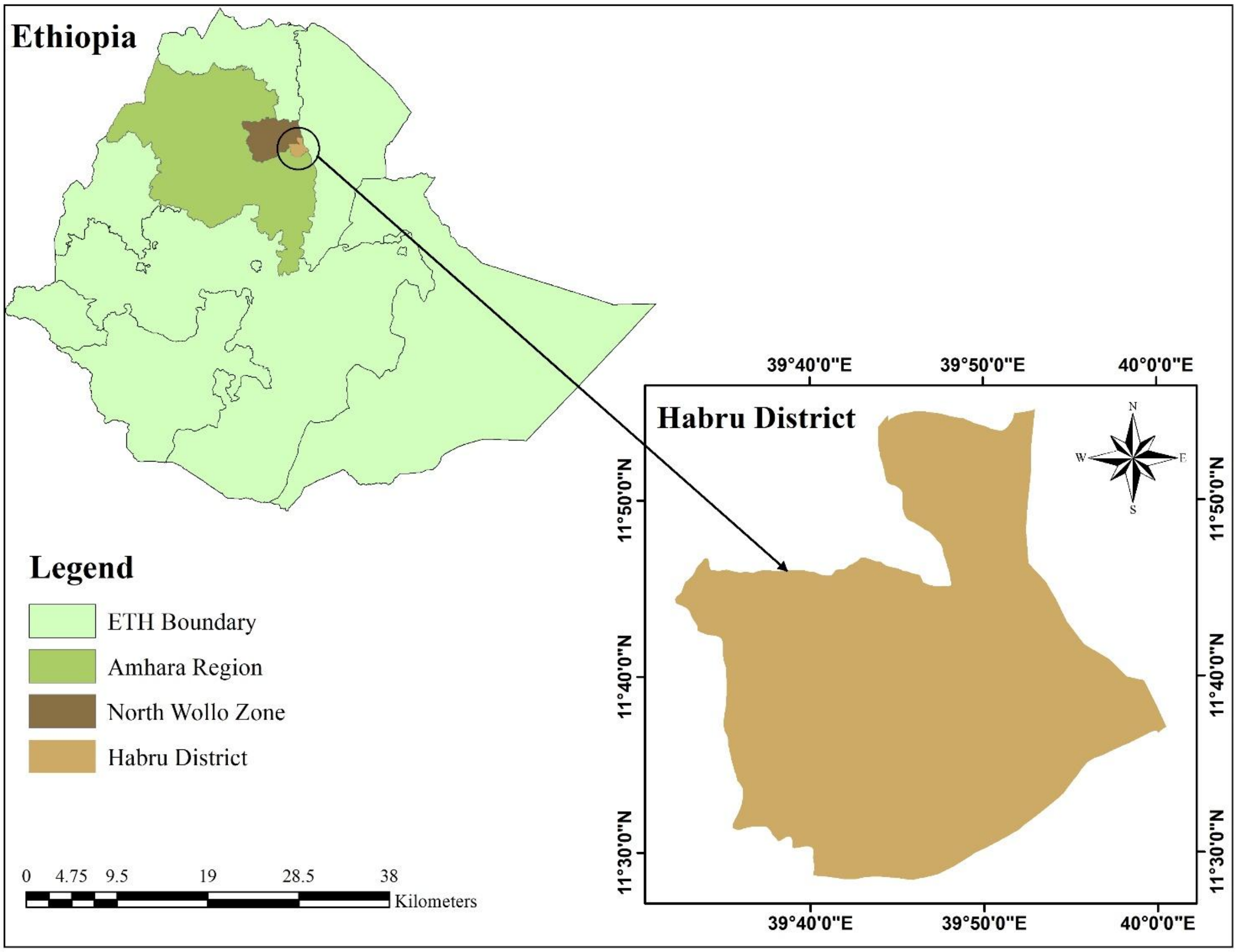

Figure 1.1: Map of Ethiopia showing North Wollo Zone of Amhara Regional State, and the location of the study area (Developed using ArcGIS 10.5)

2.2.2. Data acquisition

To assess the dynamics LULC changes, a time series of LULC datasets, derived from multispectral Landsat imagery for the years 1985, 1997, 2010, and 2021, was obtained from the United States Geological Survey (USGS) Earth Explorer website (http://earthexplorer.usgs.gov/). This step was crucial for the detailed analysis of land use and cover within the study area. During the selection process, images characterized by high resolution and minimal to no cloud interference were prioritized from the available datasets for each specified period as described in Table 1.1 (Terefe Tolessa *et al.*, 2016).

Table 1.1: Description of Landsat images and their sources

No.	Landsat Image	Sensor	Resolution or scale	Year of image acquisition	Path/row	Source
1.	Landsat 8	Operational Land Imager (OLI)	30m	2021	168/52	USGS
2.	Landsat 5	Thematic Mapper (TM)	30m	2010	168/52	USGS
3.	Landsat 5	Thematic Mapper (TM)	30m	1997	168/52	USGS
4.	Landsat 5	Thematic Mapper (TM)	30m	1985	168/52	USGS

To accurately determine the primary land use types within the study area, field observations were conducted to collect Ground Control Points (GCPs) essential for the georeferencing of images, facilitating supervised classification and enhancing the understanding of various land cover class characteristics to aid in the visual interpretation of the images. By recognizing the LULC of the study area, a comprehensive collection of 644 reference points within Habru District was conducted accordingly using a Global Positioning System (GPS) receiver, complemented by Google Earth and topographic maps of the region to support land cover classification, ground truth verification, and subsequent accuracy assessment efforts. Archival google earth images was used for the different years to find the LULC categories for the particular year. This thorough approach enabled the identification of six predominant land use land cover types in the study: water bodies, settlement areas, cultivated lands, bare lands, shrub lands, and forest lands (Table 1.2).

Table 1.2: LULC change types and their descriptions.

LULC types	Descriptions of LULC classes for which changes were detected for the period between 1985 & 2021
Bare land	Characterized by the absence of vegetation or the presence of degraded agricultural lands, bare land in the study area is predominantly located in mountainous regions, featuring bare soil and exposed rocks
Settlement area	Comprises areas designated for both rural and urban settlements, including buildings, industries, and various infrastructures. The study region consists of small villages or dispersed villages

Cultivated land	Encompasses regions allocated for the cultivation of crops (both annual and perennial), fallow plots, and certain pastures and plantations near settlements. The area is noted for its extensive fields of annual crops, plantations of perennial crops, and irrigated zones.
Forest land	Identified by dense tree coverage, these areas are dominated by tall indigenous angiosperm trees, *Eucalyptus* plantations, and coniferous trees
Shrub land	Covered with small trees, bushes, and shrubs, and occasionally interspersed with grasses, these areas are less dense than forests, with woody components being more scattered
Water body	This refers to areas covered by water, including ponds, lakes, reservoirs, rivers, and streams found within the study area.

Data concerning the major driving forces of LULC changes, their impacts, and threats to vegetation in the study area were gathered through FGD using checklist prepared for discussion and interviews with 42 key informants from the local population using semi-structured questionnaire. In total, 13 FGD were held, one was conducted at the district level, and the remaining 12 were conducted within selected kebeles of the district. Participants in each focus group comprised elders, administrative officials from both the kebele and district levels, specifically from departments concerned with natural resource and forest protection, as well as agricultural and rural development offices. Key informants, selected based on their extensive local experience of over thirty years, were purposefully chosen to provide insights into the LULC changes within the area through interviews.

2.2.3. LULC change data analysis

The investigation of land use land cover (LULC) change analysis in this study was structured around a five-step methodology, as outlined in the designated framework (Figure 1.2). In this study, GIS and remote sensing based techniques were applied to classify and analyze the LULC dynamics. This study involves image preprocessing, supervised classification, and change detection. An accurate statistical analysis of the LULC changes in the study area was conducted (Phiri and Morgenroth, 2017).

These steps provide a comprehensive approach to understanding LULC changes, enabling the identification of patterns, drivers, and impacts associated with such transformations.

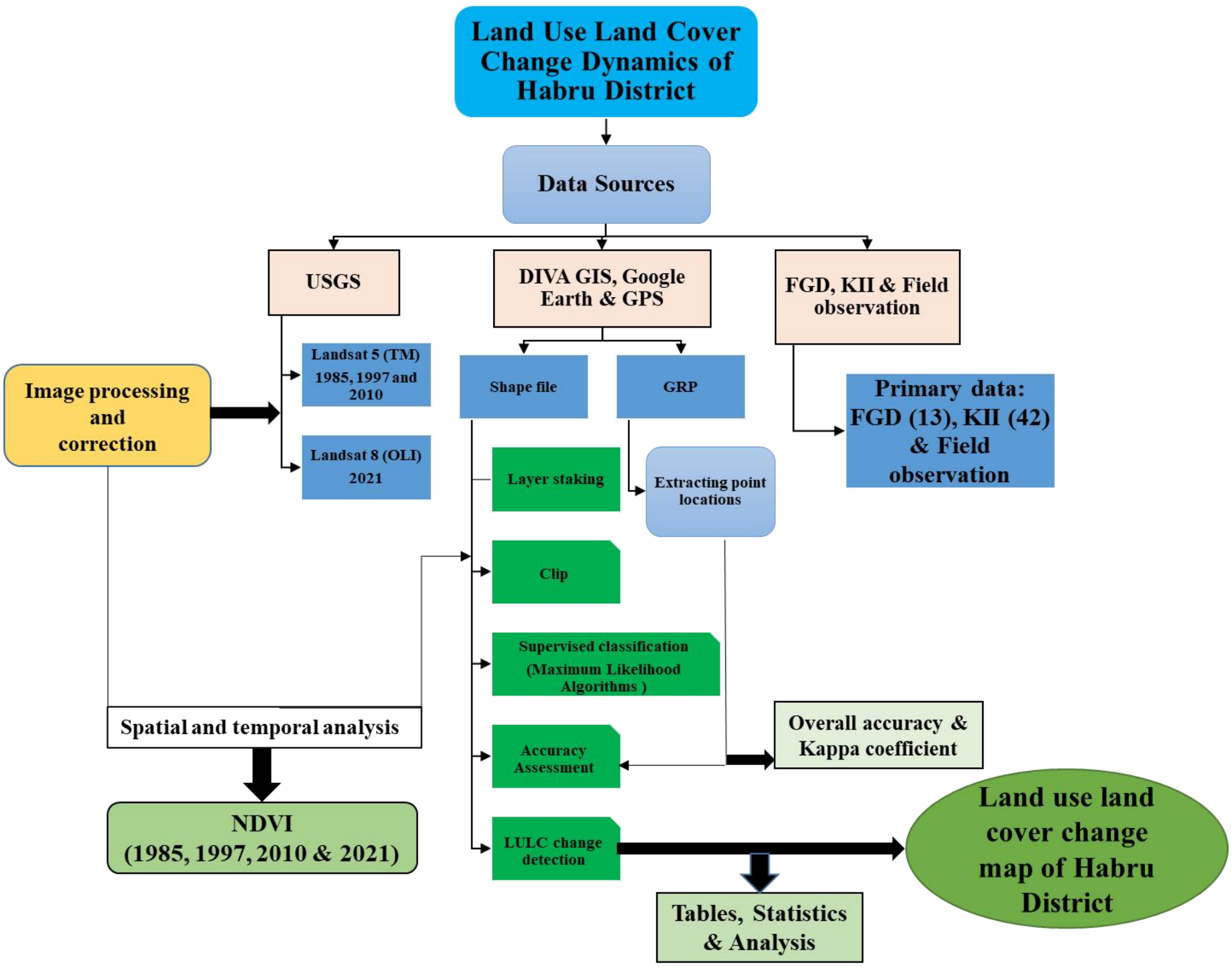

Figure 1.2: Conceptual framework for LULC change study adapted from Mehari Mariye *et al*., (2022).

Preprocessing of raster images

Preprocessing satellite imagery prior to change detection analysis is crucial for accurately reflecting the relationship between the captured data and the physical characteristics of the land surface (Coppin *et al.*, 2004). For this study, polygons representing the area of interest with various land use types were created in ArcMap 10.5 (ESRI, 2017). The digitized study area polygons were then exported as shapefiles, which were subsequently used to clip the raster images for the years 1985, 1997, 2010, and 2021. The final step involved exporting these processed images to ArcMap, where the image analysis tool was employed for the clear identification and classification of potential land use classes. This facilitated the enhancement and correction of resampled images, setting the stage for accurate change detection and analysis.

Image classification

The classification phase is aimed at categorizing the landscape into different land use and land cover classes based on the preprocessed images (Boakye *et al.*, 2008; Al-sharif and Pradhan, 2014). This study employed a supervised classification method utilizing a minimum likelihood algorithm, which relies on ground reference points for accurate categorization (Phiri and Morgenroth, 2017; Temesgen Gashaw *et al.*, 2017). Prior to the classification, the images were projected to the spatial reference coordinate systems of Adindan or UTM zone 37N to ensure spatial consistency.

Accuracy assessment

Accuracy assessment is a crucial step following the post-classification processing of classified images, essential for evaluating the fidelity of the classification to actual ground conditions. This assessment validates the extent to which the classifications reflect the real-world scenario on the ground (Temesgen Gashaw *et al.*, 2014; Congalton and Green, 2019). Without performing an accuracy assessment, the trustworthiness of the classified image results remains uncertain, limiting the reliability of any conclusions drawn from the data (Ermias Teferi *et al.*, 2013; Congalton and Green, 2019). Adhering to the recommendation of having a minimum of 50 ground reference points per class for a robust accuracy assessment (Congalton, 1991), this study utilized a comprehensive dataset of 644 ground truth samples. These samples, representing each of the five land use land cover classes, were meticulously collected through field surveys employing GPS devices with an accuracy of ±5 meters. Additionally, we used the archival google earth images for the different years to find the LULC categories for the particular year. The information was systematically integrated and analyzed in ArcGIS 10.5. The process involved comparing the classified images with the reference data using an error matrix (also known as a confusion matrix or contingency table), a method applied to quantify the accuracy of classification (Rientjes *et al.*, 2011; Adenew Taffa *et al.*, 2015). The error matrix facilitates the calculation of key accuracy metrics including the producer's accuracy, user's accuracy, overall accuracy, and the Kappa coefficient (Congalton and Green, 2019).

The formula for overall accuracy (OA) is given as:

$$\mathbf{OA} = \left(\frac{\boldsymbol{x}}{\boldsymbol{y}}\right) * \mathbf{100}$$

Where, OA represents the overall accuracy, X denotes the number of correct values in the diagonals of the matrix, and Y represents the total number of values used as reference points.

The Kappa coefficient (K) formula is expressed as:

$$\boldsymbol{K} = \frac{\boldsymbol{N}\sum_{\boldsymbol{i=1}}^{\boldsymbol{r}} \boldsymbol{xii} - \sum_{\boldsymbol{i=1}}^{\boldsymbol{r}} (\boldsymbol{xi} + \ * \boldsymbol{x} + \mathbf{1})}{\boldsymbol{N}^{\mathbf{2}} - \ \sum_{\boldsymbol{i=1}}^{\boldsymbol{r}} (\boldsymbol{xi} + \ * \boldsymbol{x} + \boldsymbol{i})}$$

Where, *K* is Kappa coefficient, *r* is the number of rows in the matrix, *xii* is the number of observations in row *i* and column *i, xi*+ are the marginal totals of row *i*, *x*+*i* are the marginal totals column *i*, and *N* is the total number of observations.

LULC change detection and rates of change

Change detection analysis is pivotal for understanding the dynamics of LULC across different time frames, offering insights into how and why these landscapes evolve. This study employs a variety of techniques to identify, delineate, and quantify the differences observed in images captured of the same area but at different times or under varying conditions (Richards, 2022). In this study, a post-classification comparison change detection method was used. This approach allows for the direct comparison of classified maps from different times, providing detailed insights into the specific nature of changes occurring between various LULC classes (Ermias Teferi *et al.*, 2013; Kflay Gebrehiwot, 2020). To quantify the extent and pace of LULC transformations for each identified class over the specified time periods, calculations were performed using the MS Excel spreadsheet program. The percentage and rate of change for each LULC category were determined using specific formulae (Rao *et al.*, 2017).

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$$\mathbf{PoC} = \frac{\boldsymbol{A2} - \boldsymbol{A1}}{\boldsymbol{A1}} * \mathbf{100}$$

$$\boldsymbol{RoC} = \frac{\boldsymbol{A2} - \boldsymbol{A1}}{\boldsymbol{A1}} * \frac{\mathbf{1}}{(\boldsymbol{T2} - \boldsymbol{T1})} * \mathbf{100}$$

Where,

RoC = Rate of change

PoC = Percentage of change of a particular LULC class

A1 = Area of the previous land use class

A2 = Area of the recent land use class

T2 = Current year

T1 = Previous year

2.2.4. Normalized difference vegetation index (NDVI)

The concept of NDVI was initially proposed by Kriegler (1969) and is calculated by taking the difference between the red and near-infrared (NIR) bands. The NDVI is a common and widely used remote sensing index used to measure the amount and vigor of vegetation on the land surface (Bhandari *et al*., 2012). NDVI is employed to assess the green density of a land area by observing the unique colors (wavelengths) of visible and near-infrared sunlight reflected by vegetation (Agenagnew Gessesse and Assefa Melesse, 2019; Huang *et al.*, 2021). The index is derived from satellite imagery, where the spectrometer or radiometric sensor of the satellite captures and stores reflectance values for red and NIR bands on two distinct channels or images (Crippen, 1990). Thus, for this study we employed the NDVI to evaluate vegetation coverage within the research area using the following formula:

$$\mathbf{NDVI} = \frac{\mathbf{NIR} - \mathbf{RED}}{\mathbf{NIR} + \mathbf{RED}}$$

where, NIR and RED represent the spectral reflectance measurements recorded in the visible red and near-infrared segments of the electromagnetic spectrum, respectively (i.e. RED = the red portion of the electromagnetic spectrum and NIR = the near infrared portion of the electromagnetic spectrum). NDVI values range from -1.0 to 1.0, where negative values represent clouds and water, values near zero indicate bare soil, and higher positive values (ranging from 0.1 to 1.0) correspond to varying degrees of vegetation density, from sparse (0.1 - 0.5) to dense green vegetation (0.6 and above).

2.3. Results

2.3.1. Land use land cover change classification

Over the course of several decades, Habru District has experienced significant alterations in its land use and land cover (LULC) composition. This analysis categorized six primary LULC types (water bodies, settlement areas, cultivated lands, bare lands, shrub lands, and forest lands) for the years 1985, 1997, 2010, and 2021 (Figure 1.3), revealing the total land area of the study district to be 1,350.4322 km² (135,043.22 hectares). The distribution and percentage change of each LULC type across the four analyzed periods are detailed in Table 1.3. For the Thematic Mapper (TM) 1985 imagery, shrub land was the predominant LULC type, encompassing approximately 965.821 km² (71.5%) of the district. Forest land and cultivated land occupied 173.955 km² (12.9%) and 165.757 km² (12.3%) of the area, respectively. Bare land and settlement areas were less prevalent, covering 41.5899 km² (3.1%) and 3.1248 km² (0.2%) of the district total area respectively, as depicted in Figure 1.3. By 1997, shrub land and cultivated land constituted the largest LULC categories, spanning 813.82 km² (60.26%) and 413.765 km² (30.64%), respectively. Forest land, bare land, and settlement areas were documented over 97.9164 km² (7.25%), 16.6302 km² (1.23%), and 5.2425 km² (0.039%) of the district, correspondingly. The smallest coverage was observed in water bodies, accounting for merely 3.0573 km² (0.23%).

Table 1.3: Areal coverage summary of different LULC classes in the Habru District from 1985 – 2021

LULC types	1985		1997		2010		2021	
	Area (Km^2)	% T. area	Area (Km^2)	% T. area	Area (Km^2)	% T. area	Area (Km^2)	% T. area
Water body (WB)	0.18	0.01	3.06	23.00	8.34	0.62	2.53	0.19
Settlement area (SA)	3.12	0.23	5.24	0.39	11.66	0.86	34.83	2.58
Cultivated land (CL)	165.76	12.27	413.77	30.64	407.96	30.21	568.85	42.12
Bare land (BL)	41.59	3.08	16.63	1.23	11.32	0.84	36.39	2.69
Shrub land (SL)	965.82	71.52	813.82	60.26	800.38	59.27	612.48	45.35
Forest land (FL)	173.96	12.88	97.92	7.25	110.77	8.20	95.36	7.06

Where % is the percentage of each class out of the total area.

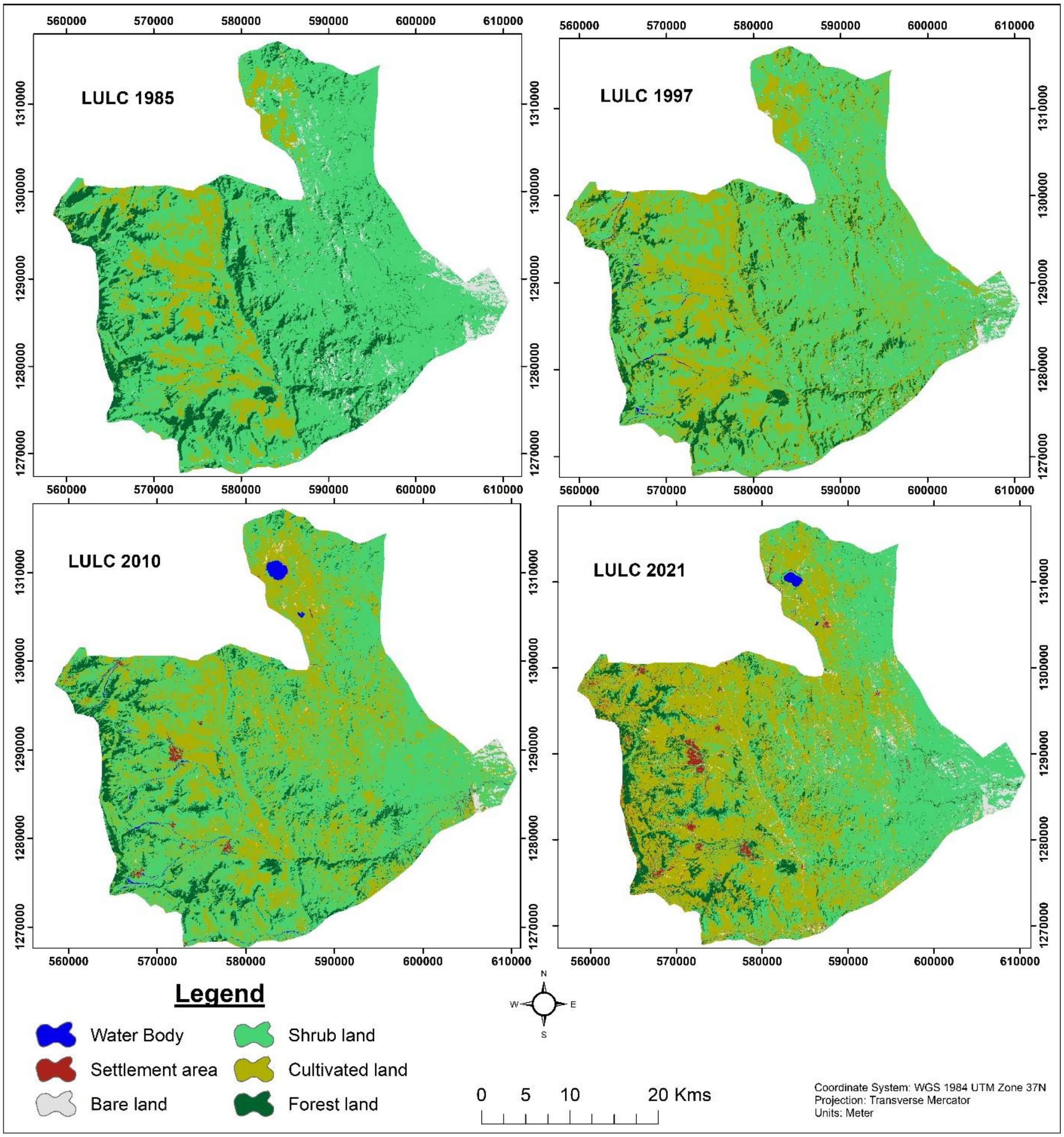


Figure 1.3: Land use land cover change map of Habru District in 1985, 1997, 2010 and 2021

The analysis of the Thematic Mapper (TM) imagery further elucidates that shrub land continues to dominate the LULC composition within the Habru District, encompassing approximately 800.383 km² (59.27%) of the district's total area. The cultivated and forested areas account for 407.963 km² (30.21%) and 110.768 km² (8.2%) of the district's landscape, respectively. Meanwhile, bare land, settlement areas, and water bodies occupy smaller fractions, comprising

11.6649 km² (0.86%), 11.3157 km² (0.84%), and 8.3367 km² (0.62%) of the area, respectively. The 2010 data highlights that shrub land and cultivated areas remain the most extensive LULC types, indicating ongoing transformations of various LULC classes into areas designated for settlements and water bodies, as detailed in Table 1.3.

The classification outcomes for the Operational Land Imager (OLI) imagery reveal that the predominant LULC categories within the Habru district remain as shrub land and cultivated land, occupying 612.4806 km² (45.35%) and 568.8486 km² (42.12%) of the district's total area, respectively. Following these, forest land, bare land, and settlement areas represent smaller portions of the district, covering 95.355 km² (7.06%), 36.3897 km² (2.69%), and 34.8273 km² (2.58%) of the total area, in that order. The minimal coverage is attributed to water bodies, covering 2.5299 km² (0.19%) of the district. The 2021 data further underscore the continued predominance of shrub land and cultivated areas, indicating ongoing shifts of other LULC classes towards settlements and water bodies, as detailed in Table 1.3 and illustrated in Figures 1.3, 1.4, and 1.6.

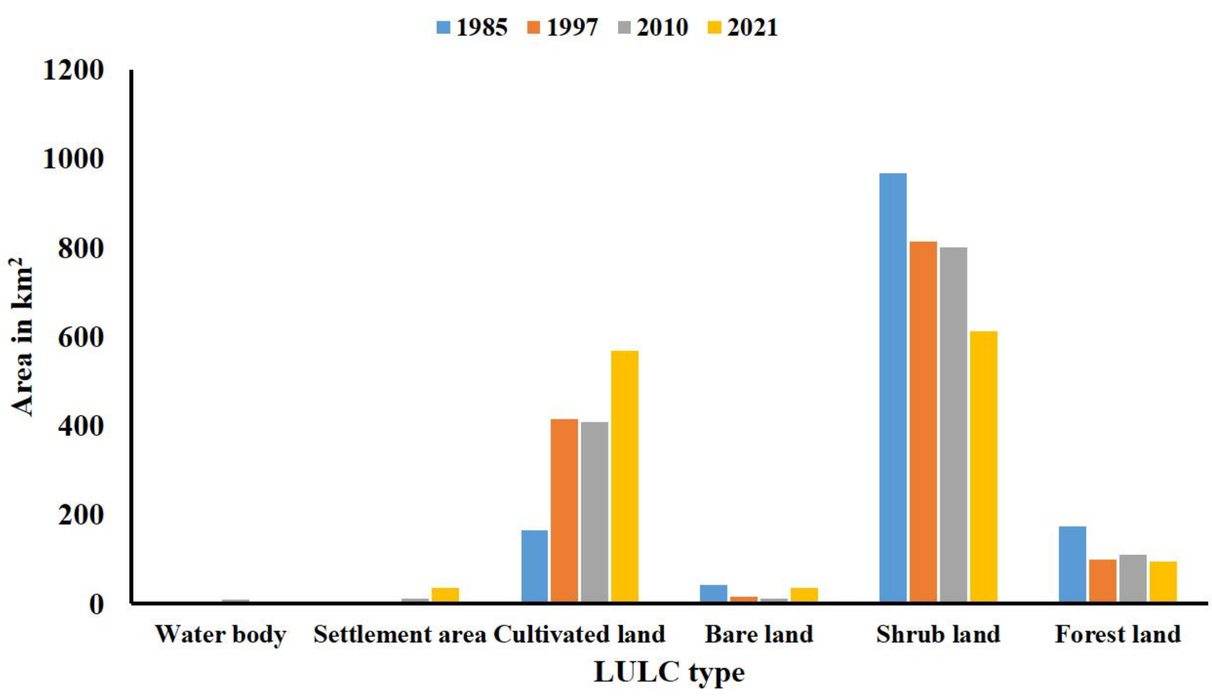


Figure 1.4: Area shifting of land use land cover classes at different periods in Habru District

2.3.2. Normalized difference vegetation index (NDVI) changes

The NDVI results for the study area from 1985 to 2021 show varying vegetation indices, indicating changes in vegetation cover and land use over time. In 1985, the area had a relatively high mean NDVI value of 0.274, which decreased by 1997 to 0.220, suggesting a reduction in vegetation cover. The lowest mean NDVI value was recorded in 2010 (0.145), indicating further decline in vegetation health or cover. However, there was a slight recovery in 2021, with the mean NDVI increasing to 0.187 (Figure 1.5).

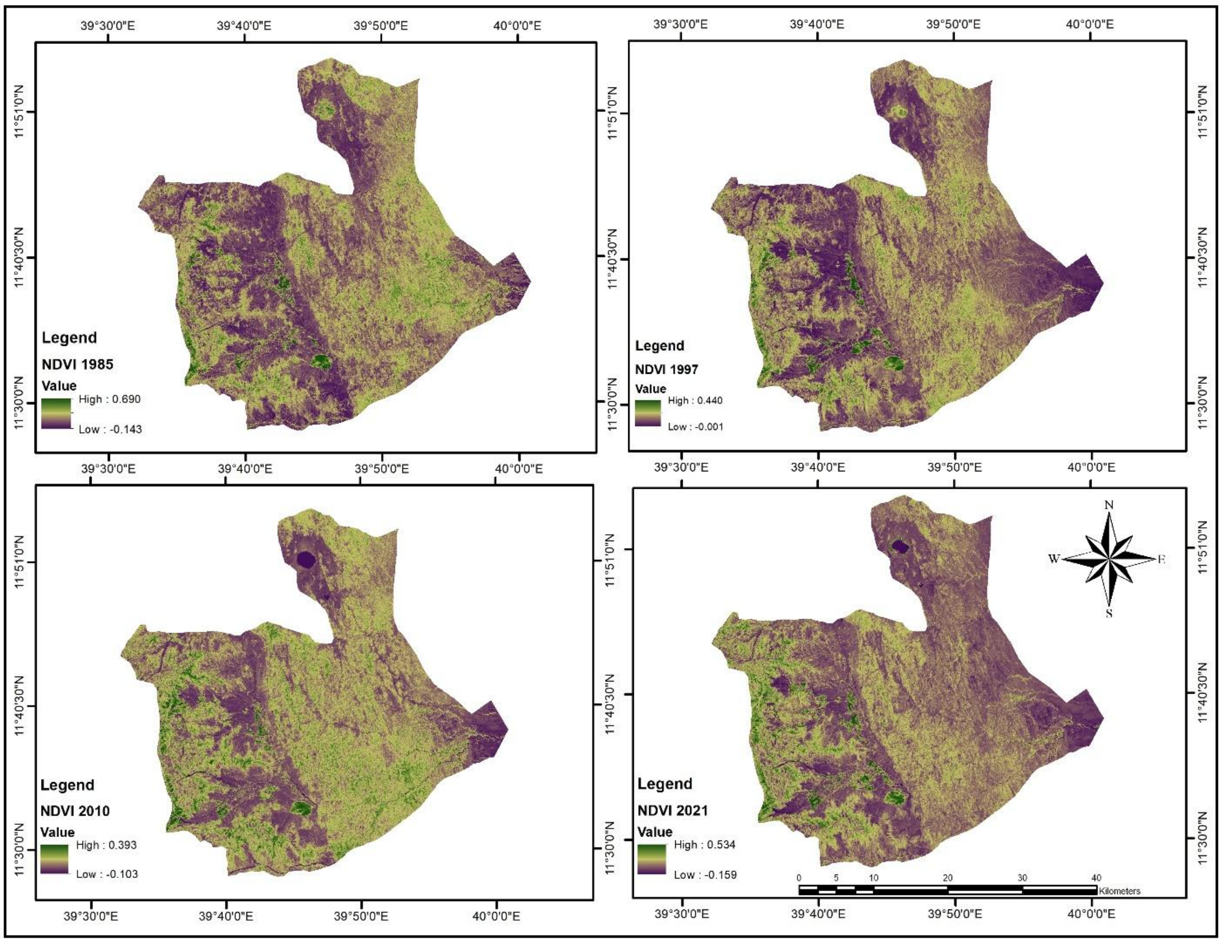
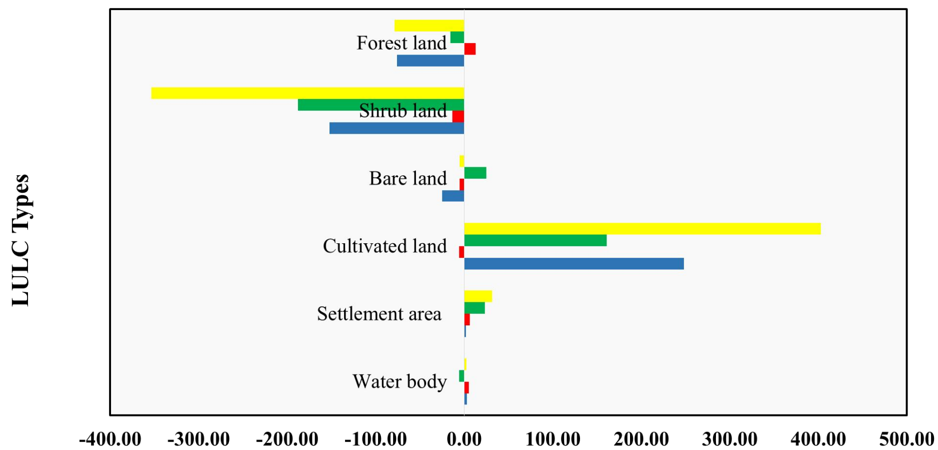


Figure 1.5: Normalized difference vegetation index (NDVI) map of the study area from 1985 - 2021



	Water body	Settlement area	Cultivated land	Bare land	Shrub land	Forest land
1985 - 2021	2.35	31.70	403.09	-5.20	-353.34	-78.60
2010 - 2021	-5.81	23.16	160.89	25.07	-187.90	-15.41
1997 - 2010	5.28	6.42	-5.80	-5.31	-13.44	12.85
1985 - 1997	2.87	2.12	248.01	-24.96	-152.00	-76.04

Change area (km^2)

Figure 1.6: Land use land cover change detection from 1985 – 2021 in Habru District

2.3.3. LULC changes and its dynamics

LULC change rates, trends and spatial extent

The analysis of land use/land cover (LULC) changes across the intervals of 1985, 1997, 2010, and 2021 highlights the extent, rate, and trends of these alterations. Between 1985 and 1997, significant shifts were observed, with the area classified as water bodies and cultivated land expanding by +1557.07% and +149.62%, respectively, indicative of substantial growth compared to the previous extents. Conversely, bare land and forest land experienced declines of -60.01% and -43.71%, respectively, reflecting a trend towards reduced coverage in these categories. Similarly, shrub land decreased by 15.74% during this period (Table 1.5), while settlement areas increased by +67.77%, demonstrating a movement towards greater development and agricultural use at the expense of natural land covers. The accuracy of the LULC classifications conducted for the study period, with overall accuracy rates of 77% (Kappa coefficient = 0.71) for 1985, 87.6% (Kappa coefficient = 0.83) for 1997, and 81% (Kappa coefficient = 0.74) for 2010, indicating a high degree of reliability in the classification results for these years. The most recent assessment for 2021 produced an even higher accuracy and Kappa coefficient of 85.9% and 0.82, respectively, underscoring the precision of the LULC classification for this period as exceptionally accurate. These findings, detailed in Tables 4-8, underscore a general trend of increased utilization of land for water bodies, cultivation, and settlements, detracting from other LULC types.

Table 1.4: Accuracy assessment of LULC change classification in Habru District (1985, 1997, 2010 and 2021)

Accuracy assessment of LULC change classification in Habru District (1985)

LULC Type	Water body (WB)	Settlement area (SA)	Bare land (BL)	Cultivated land (CL)	Shrub land (SL)	Forest land (FL)	Row total	Users accuracy %
Water body (WB)	8	0	0	0	0	0	8	100.0%
Settlement area (SA)	0	8	0	0	0	0	8	100.0%
Bare land (BL)	0	0	7	0	0	0	7	100.0%
Cultivated land (CL)	0	0	0	28	1	0	29	96.6%
Shrub land (SL)	2	3	3	6	36	1	51	70.6%
Forest land (FL)	0	0	1	0	16	21	38	55.3%
Column tot	10	11	11	34	53	22	108	
Producer accuracy %	80.0%	72.7%	63.6%	82.4%	67.9%	95.5%	**0.77**	

Note: The overall classification accuracy is **77%** whereas the overall kappa statistics 0.7

Accuracy assessment of LULC change classification in Habru District (1997)

LULC Type	Water body (WB)	Settlement area (SA)	Bare land (BL)	Shrub land (SL)	Cultivated land (CL)	Forest land (FL)	Row total	Users accuracy %
Water body (WB)	8	0	0	0	0	0	8	100.0%
Settlement area (SA)	0	9	0	0	0	0	9	100.0%
Bare land (BL)	0	0	8	0	0	0	8	100.0%
Shrub land (SL)	0	2	2	58	7	1	70	82.9%
Cultivated land (CL)	0	0	1	3	31	1	36	86.1%
Forest land (FL)	0	0	1	2	0	27	30	90.0%
Column tot	8	11	12	63	38	29	**141**	
Producer accuracy %	100.0%	81.8%	66.7%	92.1%	81.6%	93.1%	87.6%	

Note: The overall classification accuracy is **87.6%** whereas the overall kappa statistics **0.83**

Accuracy assessment of LULC change classification in Habru District (2010)

LULC Type	Water body (WB)	Shrub land (SL)	Forest land (FL)	Bare land (BL)	Cultivated land (CL)	Settlement area (SA)	Row total	Users accuracy %
Water body (WB)	11	0	0	0	0	0	11	100%
Shrub land (SL)	0	37	2	1	14	3	57	65%
Forest land (FL)	0	6	27	0	0	0	33	82%
Bare land (BL)	0	0	0	4	0	0	4	100%
Cultivated land (CL)	0	3	1	2	44	0	50	88%
Settlement area (SA)	0	0	0	0	0	10	10	100%
Column tot	11	46	30	7	58	13	133	
Producer accuracy %	100%	80%	90%	57%	76%	77%	81%	

Note: The overall classification accuracy is **81%** whereas the overall kappa statistics **0.74**

Accuracy assessment of LULC change classification in Habru District (2021)

LULC Type	Water body (WB)	Settlement area (SA)	Bare land (BL)	Shrub land (SL)	Cultivated land (CL)	Forest land (FL)	Row total	Users accuracy %
Water body (WB)	12	0	0	0	0	0	12	100%
Settlement area (SA)	0	16	0	0	0	0	16	100%
Bare land (BL)	0	1	8	0	1	0	10	80%
Shrub land (SL)	0	1	3	35	1	5	45	78%
Cultivated land (CL)	0	0	3	5	56	0	64	88%
Forest land (FL)	0	0	0	5	0	25	30	83%
Column tot	12	18	14	45	58	30	152	
Producer accuracy %	100%	89%	57%	78%	97%	83%	85.9%	

Note: The overall classification accuracy is **85.9%** whereas the overall kappa statistics **0.82**

Table 1.5: Extent and rate of LULC changes (km^2/year) between 1985 and 2021 in Habru District

LULC types	Change (1985 - 1997)		Change (1997 - 2010)		Change (2010 - 2021)		Change (1985 - 2021)		Annual rate change (1985 - 1997)		Annual rate change (1997 - 2010)		Annual rate change (2010 - 2021)		Annual rate change (1985 - 2021)	
	Area (Km2)	%a	Area (Km2)	%a	Area (Km2)	%a	Area (Km2)	%a	Area (Km2)	%b	Area (Km2)	%b	Area (Km2)	%b	Area (Km2)	%b
Water body (WB)	2.87	1557.07	5.28	172.68	-5.81	-69.65	2.35	1271.22	129.76	1557.07	13.28	172.68	-6.33	-69.65	35.31	1271.22
Settlement area (SA)	2.12	67.77	6.42	122.51	23.16	198.56	31.70	1014.54	5.65	67.77	9.42	122.51	18.05	198.56	28.18	1014.54
Cultivated land (CL)	248.01	149.62	-5.80	-1.40	160.89	39.44	403.09	243.18	12.47	149.62	-0.11	-1.40	3.59	39.44	6.76	243.18
Bare land (BL)	-24.96	-60.01	-5.31	-31.96	25.07	221.59	-5.20	-12.50	-5.00	-60.01	-2.46	-31.96	20.14	221.59	-0.35	-12.50
Shrub land (SL)	-152.00	-15.74	-13.44	-1.65	-187.90	-23.48	-353.34	-36.58	-1.31	-15.74	-0.13	-1.65	-2.13	-23.48	-1.02	-36.58
Forest land (FL)	-76.04	-43.71	12.85	13.13	-15.41	-13.91	-78.60	-45.18	-3.64	-43.71	1.01	13.13	-1.26	-13.91	-1.26	-45.18

Where, a Percentage change in the component; b Percentage of the annual rate of change in each class.

LULC change matrix

The analysis of change matrices for each examined period provided insights into the origins and destinations of significant LULC transformations. The matrices reveal that an area of approximately 11.48 hectares, designated as water body in 1985, retained its classification by 1997, as detailed in Table 1.6. Conversely, the data indicate a shift in land use by 2021, with about 8.17 hectares initially categorized under other LULC types in 1985 transitioning to cultivated land, and approximately 4.49 hectares evolving into bare land, as outlined in Table 1.9. Moreover, a significant conversion was observed in the extent of shrub land and forest cover from 1985 to 2021. Specifically, around 37,845.97 hectares of shrub land and 5,225.63 hectares of forest land had been repurposed as cultivated land by 2021, highlighting substantial shifts in land utilization towards agricultural activities.

Table 1.6: LULC conversion matrix of the Habru District from 1985 - 1997

		LULC Type in 1997 (ha)						
		WB	**SA**	**BL**	**CL**	**SL**	**FL**	**Total**
LULC Type in 1985 (ha)	**WB**	**11.48**	0.02	0.00	2.05	0.99	0.00	14.55
	SA	1.31	**7.66**	0.66	36.42	194.73	0.07	240.85
	BL	0.01	96.01	**1096.14**	442.51	2320.11	0.24	3955.02
	CL	5.50	10.49	9.11	**12874.78**	3446.98	21.37	16368.25
	SL	272.55	294.26	461.47	21343.95	**73658.39**	1335.65	97366.27
	FL	2.14	17.76	7.53	5476.78	3142.05	**8400.28**	17046.53
	Total	292.99	426.21	1574.90	40176.48	82763.25	9757.62	134991.45

Where: Bare land (BL), Cultivated land (CL), Forest land (FL), Settlement area (SA), Shrub land (SL) and Water body (WB)

Table 1.7: LULC conversion matrix of the Habru District from 1997 - 2010

		LULC Type in 2010 (ha)						
		WB	**SA**	**BL**	**CL**	**SL**	**FL**	**Total**
LULC Type in 1997 (ha)	**WB**	**133.02**	8.09	0.00	149.46	1.54	0.86	292.97
	SA	3.11	**38.94**	12.31	122.49	246.74	2.69	426.28
	BL	0.80	18.47	**622.14**	376.59	556.13	0.09	1574.22
	CL	272.36	591.60	389.37	**54302.27**	25459.54	1743.65	82758.79
	SL	370.85	345.54	67.31	23555.22	**13460.81**	2375.61	40175.34
	FL	13.44	33.78	0.00	2866.00	123.61	**6721.03**	9757.86
	Total	793.58	1036.42	1091.14	81372.02	39848.37	10843.92	134985.46

Where: Bare land (BL), Cultivated land (CL), Forest land (FL), Settlement area (SA), Shrub land (SL) and Water body (WB)

Table 1.8: LULC conversion matrix of the Habru District from 2010 - 2021

		LULC Type in 2010 (ha)						
		WB	**SL**	**FL**	**BL**	**CL**	**SA**	**Total**
LULC Type in 2021 (ha)	**WB**	**248.89**	130.56	17.67	59.15	319.70	17.70	793.67
	SL	2.91	**44989.90**	3133.47	1660.28	29628.02	1959.47	81374.05
	FL	0.00	2851.52	**5861.28**	2.88	2074.16	54.94	10844.78
	BL	0.00	497.05	1.50	**392.22**	196.08	4.37	1091.22
	CL	0.02	12683.55	398.30	1201.91	**24841.82**	722.33	39847.93
	SA	0.00	278.66	10.98	65.10	237.52	**444.19**	1036.45
	Total	251.81	61431.24	9423.21	3381.55	57297.30	3203.00	134988.11

Where: Bare land (BL), Cultivated land (CL), Forest land (FL), Settlement area (SA), Shrub land (SL) and Water body (WB)

Table 1.9: Major LULC conversions of the Habru district from 1985 - 2021

From Class	**To class**	**Area change from 1985 – 2021 (ha)**
Water body	Settlement area	0.36
	Bare land	4.49
	Shrub land	0.58
	Cultivated land	8.17
	Forest land	0.09
Settlement area	Bare land	10.01
	Shrub land	44.83
	Cultivated land	169.87
	Forest land	2.00
Bare land	Water body	5.19
	Settlement area	47.72
	Shrub land	2407.52
	Cultivated land	869.77
	Forest land	7.58
Cultivated land	Water body	0.99

	Settlement area	564.64
	Bare land	135.33
	Shrub land	2207.91
	Forest land	279.84
Shrub land	Water body	215.66
	Settlement area	2367.20
	Bare land	2579.28
	Cultivated land	37845.97
	Forest land	3040.20
Forest land	Water body	29.11
	Settlement area	208.57
	Bare land	34.61
	Shrub land	5455.61
	Cultivated land	5225.63

2.3.4. Major drivers of LULC changes in Habru District

Analysis of proximate factors influencing LULC change revealed that expansion of cultivated land and increased settlement areas were the primary drivers. These factors ranked first and second, respectively, followed by charcoal production, firewood collection, and tree cutting for various purposes. Furthermore, the investigation of underlying causes identified population growth/pressure and land tenure insecurity as the most significant drivers (ranking first and second, respectively). These were followed by economic crises (poverty and unemployment) and climate change (precipitation variability) (Table 1.10).

Table 1.10: Major drivers of LULC change in the study area

Category	Driver	No. of response among 42 KI's	Percentage	Rank
Proximate drivers	Cultivated land expansion	35	83.33	1
	Settlement area expansion	28	66.67	2
	Charcoal production	19	45.24	3
	Firewood collection	15	35.71	4
	Tree cutting	11	26.19	5
Underlying causes	Population growth/pressure	32	76.19	1
	Economic challenge (poverty and unemployment)	29	69.05	3
	Climate change (precipitation variability)	22	52.38	4
	Land tenure insecurity	30	71.43	2

2.4. Discussion

This study investigated the biophysical and sociocultural factors driving land use land cover (LULC) changes in Habru District. Our core objective was to identify, classify, and quantify the extent of these changes. The study utilized change detection techniques, following the framework established by Hoffer (1978), to investigate variations over time in spectral responses. These variations serve as indicators of modifications in the spectral attributes of vegetation or different land cover categories throughout the research area. These methods enabled us to pinpoint specific LULC transitions that occurred between analyzed periods (Shiferaw Abate, 2011).

This study identified six primary LULC classes in Habru District: water bodies, settlement areas, bare land, cultivated land, shrubland, and forest area. This spatial distribution suggests significant transformations within the district over at least the past 36 years. Shrubland constituted the dominant LULC class, covering approximately 71.52% of the total area in 1985 and decreasing to 45.32% in 2021. Kappa statistics were employed to assess the classification accuracy, following the approach established by Forkuor and Cofie (2011). This method evaluates both the completeness and correctness of the LULC classification scheme. The accuracy assessment for the most recent data point (2021 LULC classification) achieved an excellent rating. Similar findings were reported for LULC dynamics in Ethiopia's Awash River Basin, where overall accuracy and kappa coefficient values exceeded 88% and 0.83, respectively, for the years 1988, 2002, and 2018 (Mahtsente Tadese *et al.*, 2020). This study also revealed significant LULC changes within Habru District over the 36-year period (1985-2021). Water bodies, settlement areas, and cultivated land exhibited substantial areal increases of 1271.2%, 1014.54%, and 243.18%, respectively. Conversely, forest land, shrubland, and bare land experienced a decline in coverage, decreasing by 48.18%, 36.58%, and 12.5%, respectively, during this timeframe, indicating a transformation to other LULC classes.

This study identified a link between the expansion of cultivated land, and settlements area as a major proximate driver of LULC change in the study area, potentially leading to natural resource degradation and severe soil erosion. Additionally, charcoal production and firewood collection, common livelihood practices in rural Ethiopia, likely contribute to these LULC changes (Solomon Gebreyohannis *et al.*, 2014; Temesgen Gashaw *et al.*, 2017; Gebeyehu Abebe *et al.*, 2022). Our findings suggest that over half of the shrubland cover in 1985 remained relatively stable, constituting around 60% of the total area in both 1997 and 2010. However, by 2021, a significant shift occurred, with shrubland and cultivated land together encompassing approximately 85% of the total area.

The Normalized Difference Vegetation Index (NDVI) is a crucial metric for monitoring vegetation health and its response to climate change, utilizing light absorption and reflection properties to evaluate vegetation density and condition. NDVI values range from -1 to +1, where higher values indicate healthier vegetation (Choubin *et al.*, 2019; Faisal *et al.*, 2020; Hu *et al.*, 2023). Our analysis revealed that from 1985 to 2021, the NDVI measurements for the study area displayed variable vegetation indices, signifying changes in vegetation cover and other land use patterns. The mean NDVI value started at 0.274 in 1985, fell to 0.220 by 1997, and reached its lowest at 0.145 in 2010, denoting a reduction in vegetation health. A slight improvement was noted in 2021, with the mean NDVI value increasing to 0.187. These fluctuations in NDVI values could be attributed to various factors such as deforestation, urbanization, or natural vegetation regrowth patterns, emphasizing the dynamic nature of land use and land cover changes in the area.

Classification accuracy was assessed using overall accuracy and kappa coefficient, incorporating both user's and producer's accuracy for each LULC class (Ehsan and Kazem, 2013). Kappa interpretation follows established criteria: values below 0.4 indicate poor agreement, 0.4-0.7 indicate good agreement, and above 0.75 indicate excellent agreement (Ismail and Jusoff, 2008). Our analysis result revealed that, the overall accuracies of 77% (kappa = 0.71), 87.6% (kappa = 0.83), and 81% (kappa = 0.74) for the years 1985, 1997, and 2010, respectively. The most recent image (2021) achieved an overall accuracy of 85.9% and a kappa coefficient of 0.82, signifying excellent classification according to established benchmarks. These results align with findings by (Reis, 2008; Mahtsente Tadese *et al.*, 2020), who reported overall accuracies exceeding 86%. Therefore, the kappa statistics in this study demonstrated strong agreement for the 2021 classification, and the overall accuracy across all years justifies further analysis of LULC changes.

Confirming prior research in Ethiopia's northwestern highlands (Gete Zeleke and Hurni, 2001) and Gubalafto District (Gebeyehu Abebe *et al.*, 2022), which identified forest cover and grazing land as most susceptible to LULC changes, this study observed a similar trend. During the 1985-2021 period, cultivated land and settlement areas expanded at the expense of natural forest land and shrubland. Our findings align with other studies in Ethiopia that documented significant forest and woodland conversion to cultivated lands. For instance, Mengistie Kindu *et al.* (2013) reported such a conversion in Munessa-Shashemene (south-central highlands) from 1973 to 2012. Similar agricultural land expansion, sourced from grazing and shrublands, was observed in the north Gojjam sub-basin (Alelgn Ewunetu *et al.*, 2021) and the Jedeb watershed (Ermias Teferi *et al.*, 2013) for the periods 1986-2017 and 1972-2009, respectively.

Echoing trends across Ethiopia, focus group discussion (FGD) participants in Habru District highlighted population growth as a major environmental concern. They perceived its continuous rise to have a devastating impact on the local environment. This aligns with findings from Gebeyehu Abebe *et al.* (2022) who identified population pressure as a key driver of land-use/land-cover (LULC) changes in the adjacent Gubalafto District. Key informant (KI) interviews and FGDs further revealed that regime changes and land tenure insecurity were perceived by participants as significant factors influencing LULC changes in the study area. Informants contrasted the current situation with the Dergue regime (1974-1991), when large-scale afforestation efforts were implemented across Habru. They attributed positive environmental changes during that period, such as reduced erosion and improved wildlife habitat, to the increased tree cover.

This study also highlights the ongoing challenge of deforestation in Habru District. While large-scale afforestation efforts were undertaken during the Dergue regime (1974-1991), these gains were subsequently undermined by unsustainable resource use practices in the early 1990s. These practices included clearing trees for construction, settlements, firewood, and charcoal production. This pattern of deforestation re-emerged during the recent conflict (November 2020 – November 2022) in northern Ethiopia between TPLF and the Ethiopian government, as the local people took advantage of the lack of control to cut trees for construction, firewood, charcoal making and cultivated land expansion. Similar observations of deforestation due to political instability from the adjacent Gubalafto District were reported by Gebeyehu Abebe *et al.* (2022).

While our findings highlight drivers of LULC changes in Habru District, it's crucial to acknowledge the dependence of local livelihoods on activities such as firewood collection, charcoal production, and mixed farming systems. Key informants and focus group discussion participants highlighted that, between 2020 and 2022, there has been an increase in the exploitation of forest resources for firewood and charcoal production and sales in both rural and urban areas. This aligns with observations made by Alelgn Ewunetu *et al.* (2021) in the north Gojjam sub-basin and in Gubalafto District, who reported similar trends in these regions of Ethiopia (Gebeyehu Abebe *et al.*, 2022).

2.5. Conclusion

This study employed digital image processing techniques to quantify LULC changes in Habru District, Ethiopia, between 1985 and 2021. The analysis revealed significant LULC transformations over the study period. Six LULC classes were identified and mapped from Landsat imagery (1985, 1997, 2010, 2021): water body, settlement area, cultivated land, bare land, shrub land, and forest land. Notably, cultivated land (403.09 km^2 increase, +243.18%), settlement area (31.70 km^2 increase, +1014.54%), and water body (2.35 km^2 increase, +1271.22%) exhibited substantial expansion during 1985-2021. Conversely, shrub land (-353.34 km^2, -36.58%) and forest land (-78.6 km^2, -45.18%) experienced significant reductions. The change in bare land was negligible. Population growth and economic challenges are likely the primary drivers of cultivated land and settlement area expansion in Habru District. The observed decline in forest and shrub land cover points towards a potential combined influence of human activities, climate change, and agricultural expansion. Our findings further suggest that human activities, particularly agricultural practices involving shrubland clearing for cultivation and settlement expansion, are key factors shaping landscape dynamics in the study area. This research demonstrates the effectiveness of remote sensing and associated techniques in monitoring LULC change patterns.

Overall, to counteract the negative consequences of LULC changes in Habru District, a multifaceted approach is necessary. This includes implementing sustainable land use planning and management practices, introducing biodiversity conservation measures, and exploring alternative livelihood strategies for local communities. The involvement of experts in biodiversity conservation, natural resource management, and ecology is crucial for developing a sustainable land use plan and effectively implementing conservation practices. By prioritizing these actions, the essential ecosystem functions of Habru District can be preserved for future generations.

CHAPTER THREE

3. Woody Species Composition, Structure and Plant Community Analysis of Gerado, Micha and Mekelet Forest Patches in Habru District and its Environs, North Wollo Zone, Amhara Region, Ethiopia

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Woody Species Composition, Structure and Plant Community Analysis of Gerado, Micha and Mekelet Forest Patches in Habru District and its Environs, North Wollo Zone, Amhara Region, Ethiopia

Abstract

Ethiopia's vegetation is increasingly endangered due to various anthropogenic activities coupled with natural factors leading to the degradation and fragmentation of forest resources. To plan an effective conservation strategy, and take restoration measures, this study conducted analysis and composition of the woody species of Gerado, Micha and Mekelet forest patches in North Wello Zone of Ethiopia. In these forest patches, trees or shrubs having ≥ 2.5 cm diameter at breast height and ≥ 2 m height were collected from a total of 95 plots (20 x 20 m^2) laid systematically along elevation gradients. The Shannon-Wiener diversity index was used to quantify species richness and evenness of the forest patches. The identification of plant community types and the analysis of the relationship between these types and environmental variables were conducted using agglomerative hierarchical cluster analysis and Redundancy Analysis (RDA) within the R software framework. A total of 55 woody plant species belonging to 46 genera and 31 families were recorded from the three forest patches. The highest species richness was recorded for the Fabaceae (12 species) followed by the Anacardiaceae, Euphorbiaceae and Malvaceae (3 species each). Four plant community types ($R = 0.4703, P \leq 0.001$) significantly different from each other were identified. The RDA results showed that altitude, slope, and tree cutting had significant impacts ($p \leq 0.05$) on the composition and distribution of community types. The important value index result indicated that, *Dodonaea viscosa* subsp. *angustifolia*, *Olea europaea* L. subsp. *cuspidata* and *Vachellia sieberiana* are the species that commonly dominate as ecologically important across the three forest patches. The study also identified anthropogenic disturbances such as tree cutting, firewood collection, charcoal production, agricultural expansion, grazing, and browsing by domestic animals as important factors that affect the species distribution and vegetation structure of the forest patches. This study reveals that existing management efforts in the Gerado, Micha, and Mekelet forest patches may be inadequate to fully mitigate anthropogenic disturbances affecting woody species composition and population structure. It suggests the need for adopting of integrated, community-based forest conservation practices, specifically designed for each forest's unique ecological and socio-economic context to enhance ecosystem resilience and sustainability.

Key Words: Anthropogenic Disturbances, Conservation, Forest Patch, Plant Community, Species Composition

3.1. Introduction

Forest ecosystems play multiple roles at local, national, and global levels through the provision of various goods and services including food, medicine, ecological and sociocultural advantages (Cheng *et al.*, 2017). Tropical forests are known for their high levels of biodiversity, which support numerous living species and constitute the complex webs of life (Pillay *et al.*, 2022). Ethiopia is characterized by a wide range of ecological, edaphic, and climatic conditions that account for the wide diversity of biological resources, both in terms of flora, faunal and microbial wealth (Vavilov, 1951; Westphal, 1974). Its complex topography and environmental heterogeneity foster a wide array of life forms (Zerihun Woldu *et al.*, 1999; Gete Zeleke, 2003). The country is in tropics with diverse vegetation types, including forests and woodland vegetation (Mengesha Asefa *et al.*, 2020). Even though the diverse ecosystems of Ethiopia have endowed the country with various biological wealth of plants and animals, for a variety of reasons, these resources are under severe threat (Ermias Aynekulu *et al.*, 2012). While population growth and associated anthropogenic activities such as extractive uses, burning, and land conversion are significant drivers of rapid changes in natural forests (Backéus *et al.*, 2006), natural factors also contribute considerably to the changes. Moreover, Ethiopia's biodiversity is under threat from a range of factors, including habitat conversion, unsustainable resource utilization, invasive species, the replacement of indigenous varieties and breeds, climate change, pollution, as well as lack of awareness and coordination among stakeholders (Teklu Gebretsadik, 2016; Mengesha Asefa *et al.*, 2020). These threats will have detrimental effects on the country's ecosystems and the biological diversity therein.

Furthermore, rapid population growth accompanied by agricultural land expansion, overgrazing, and uncontrolled timber and fuel wood extractions are primary drivers of deforestation in Ethiopia (Edwards *et al.*, 1999; Taye Bekele *et al.*, 1999; Yohannes Kidane *et al.*, 2012; Tsehaye Gebrelibanos and Mohammed Assen, 2015; Alemayehu Urgesa *et al.*, 2016). Consequently, the country's northeast and northwest regions retain small remnants of natural forests that continue to face increasing human-induced pressures. Within the Wollo Floristic region, the Gerado, Micha, and Mekelet forests patches, are experiencing the aforementioned anthropogenic pressures. Thus, understanding the ecological information on the floristic composition and distribution of plant species would be vital for proper conservation and development planning, and sustainable management of these forest patches. Given this, the present study aims to examine the status of the floristic composition, vegetation structure and plant community types along with environmental and anthropogenic disturbance factors of Gerado, Micha and Mekelet forest patches in the North Wollo Zone of Amhara region, Ethiopia.

3.2. Materials and methods

3.2.1. Description of the study area

This study was conducted in Habru and Gubalafto Districts, located in the North Wollo Zone of Amhara National Regional State, Ethiopia (Figure 2.1). Mersa town is the center of Habru District, which is 88 km north of Dessie town and 491 km north of Addis Ababa, the Ethiopian capital, 406 km east of Bahirdar (Amhara Region's capital), 30 km northeast of Woldiya (North Wollo Zone capital) (HDARDO, 2019). The grid references for Habru District come within 11°35'0"-11°55'0"N and 39°30'10"- 40°10'0"E with an altitudinal range of 1430-2800 masl. It is bordered to the south by the Mille River, separating it from the South Wollo Zone. Gubalafto District is situated to the west of Habru District, and the Alawuha River delineates the northern boundary, separating it from Kobo District. The eastern border is defined by the Afar Region. The agroecological zone of Habru District belongs to the warmest zone, which is locally known as Kolla (warm semi-arid) zone covering 56.5%, the cold climate zone locally known as Dega (cool and humid) covering 3.5% and medium/intermediate zone, which is locally known as Weina dega (cool sub-humid) and covers 40% of its total area (HDARDO, 2019; EBI, 2022).

The principal town in Gubalafto District is Woldia, which lies approximately 506 km northeast of Addis Ababa, situated on the main road connecting it to the towns of Mekele, Dessie, Bahir Dar, and Lalibela. It consists of 34 kebeles, and it is situated between 39° 12' 9"–39° 45'58" East and 11° 34' 54"–11° 58' 59" North with an altitudinal range of 1100-3700 masl. The district is classified into four agro-ecological zones namely, Kolla (1100–1500 m.a.s.l), Woina dega (1500–2300 m.a.s.l), Dega (2300–3200 m.a.s.l.), and Wurch or cold and moist (peaks, 3200–3700 m.a.s.l.) (Getnet Chekole, 2017; EBI, 2022). The average yearly temperature of the study area is 20.1 °C, with monthly temperatures ranging from a minimum of 10.6 °C to a maximum of 30.8 °C. The study area receives a bimodal pattern of rainfall of about 1045 mm/year in which high and low rainfall was received during Kiremt (June to October) and Belg (March and April) respectively (ENMA, 2019; Mulugeta Alemu *et al.*, 2024).

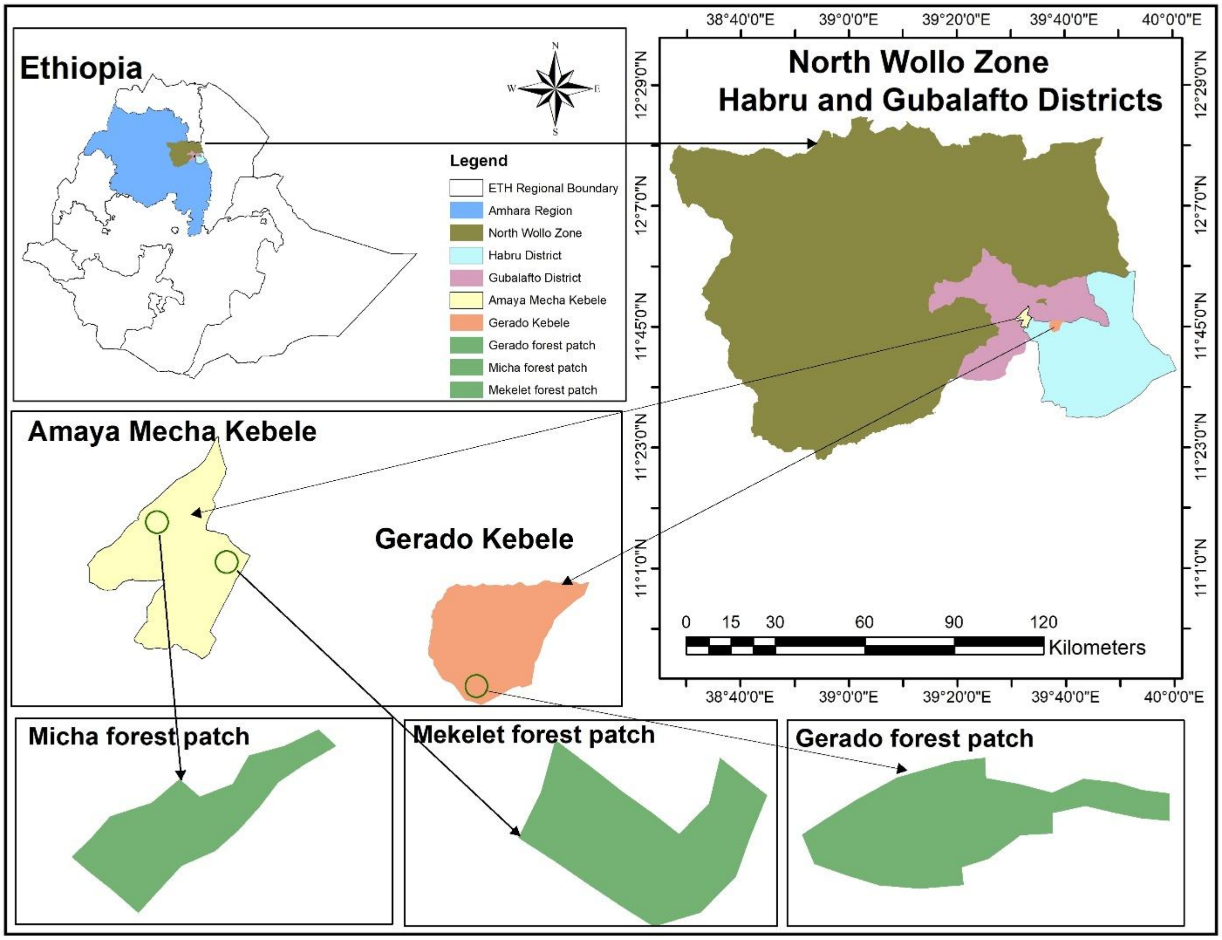

Figure 2.1: Map of Ethiopia showing the location of the three forest patches in Habru District and its environs, North Wollo Zone of Amhara Regional State (Developed using ArcGIS 10.5)

3.2.2. Sampling design and vegetation data collection

For the study, a systematic sampling approach based on Ellenberg and Mueller-Dombois (1974), complemented by the Braun-Blanquet scale as modified by Van der Maarel (1979) was used to estimate the cover-abundance value of all woody plants. A total of 95 quadrats (400 m^2) were sampled from all forest patches along elevation gradients. A total of 26 transect lines were laid across the forest patches. The number of sampling plots, distance between plots and distance between transect lines varied per forest patches based on the forest cover, altitudinal difference and habitat variability of the forest patches following the methods recommended by Kent and

Coker (1992). All line transects and plots were laid, and topographic variables like altitude, latitude, and longitude were recorded for each plot using GPS.

The percent cover of trees and shrubs was visually estimated. The scientific and local names, abundance and habits of each woody species were recorded in each plot. Within each plot, we recorded the diameter and height for woody species, measuring the diameter of trees and shrubs at breast height ≥2.5 cm using a tree caliper. Plant heights were determined using a calibrated stick for trees and shrubs. Environmental variables such as altitude, longitude, latitude, and aspect of each quadrat, were measured using GPS. For analytical clarity, aspect measurements were codified in accordance with the established system (Zerihun Woldu *et al.*, 1989). This coding translated the cardinal and intercardinal directions into numerical values: North (N) as 0, Northeast (NE) as 1, East (E) as 2, Southeast (SE) as 3, South (S) as 4, Southwest (SW) as 3.25, West (W) as 2.5, and Northwest (NW) as 1.25. The impact of anthropogenic disturbance (cutting, charcoal production, agricultural expansion, firewood collection, grazing and browsing) was recorded in the quadrats using a 0–3 subjective ordinal scale as 3 = heavy, 2 = moderate, 1= low and 0 = nil (Zerihun Woldu and Backéus, 1991; Kumelachew Yeshitela and Tamrat Bekele, 2002; Fitsum Temesgen *et al.*, 2021). Voucher specimens of plants were collected, and the preliminary identification was executed using manuals in the field and the collected specimens were pressed, dried, and brought to the National Herbarium (ETH) of Addis Ababa University, deep frozen, and determinations were undertaken. The determination was performed using the taxonomic keys of the Flora of Ethiopia and Eritrea, followed by comparisons with authenticated specimens in the ETH, and finally confirmed by a taxonomic expert at Addis Ababa University. Following identification, each specimen was assigned a voucher number and detailed information was recorded, including family, species, vernacular names, collection date, and collection site. These vouchered specimens were then deposited at the ETH for future reference.

3.2.3. Data analysis

Plant diversity analysis: The floristic diversity of the study area was measured using the Shannon-Wiener diversity index to quantify species richness and evenness (Kent and Coker, 1992). The Shannon-Wiener diversity index was calculated as follows:

$$\mathbf{H}' = -\sum_{i=1}^{s} pi\ln pi$$

Where, **H'** = Shannon-Wiener Diversity Index; **S** = The number of species; **Pi** = the proportion of individuals or the abundance of the i^{th} species expressed as a proportion of total cover; **In** = logbase_n

The Shannon's evenness or equitability of the woody species was measured as:

$$J = \frac{H'}{H'max}$$

Where: **J** = Evenness, **H' =** Shannon-Wiener Diversity Index; **H' max =** Ins, where s is number of species

Plant community classification: Plant community types of forest patches were classified using Agglomerative hierarchical clustering analysis using R statistical software version 4.2.2 (R Core Team, 2022). Considering all forest patches as one, four plant community types were identified using Ward's method of hierarchical clustering technique as recommended by Kent and Coker (1992). In this study, the optimal number of clusters was determined using the elbow method. This method involves plotting the within-group sum of squares (WSS) against the number of clusters and identifying the point where the rate of decrease sharply changes, known as the 'elbow point. For our dataset, the elbow point was observed at four clusters, indicating this as the optimal number for our k-means clustering analysis (Zerihun Woldu, 2017). Synoptic values were computed as the result of average cover-abundance values of species and their frequency in a specific community type. Thus, two characteristic species with high synoptic cover-abundance values were used to name the plant community types (Kent and Coker, 1992). Floristic similarity analysis between forest patches of the study area were computed using Sorensen's similarity coefficient as follows:

$$Ss = \frac{\mathbf{2a}}{\mathbf{2a + b + c}}$$

Where: Ss = Sorensen's similarity coefficient; a = number of species common to both forest patches compared; b = number of species in one forest patch; c = number of species in the other forest

Detrended Correspondence Analysis (DCA) was utilized to discern the appropriateness of Canonical Correspondence Analysis (CCA) or Redundancy Analysis (RDA) for examining the interplay between plant communities and environmental variables (Šmilauer and Lepš, 2014). Given the first DCA axis lengths (DCA1: 2.7942), which indicates that the data likely exhibits linear rather than unimodal species responses to the gradients (Table 2.4). Therefore, the outcomes of DCA indicated that RDA was the most suitable method, subsequently guiding the exploration of associations between all documented plant species within the communities and various environmental factors, including altitude, slope, aspect, and anthropogenic disturbances. Additionally, the Adonis 2 test was utilized to further investigate the impact of these environmental factors on the distribution of plant communities (Table 2.5).

Anthropogenic disturbance: The impact of disturbance was quantified using the variable scores recorded in each sample plot for all forest patches. The degree of disturbance was estimated based on the average score value of anthropogenic factors (Zerihun Woldu and Backéus, 1991; Kumelachew Yeshitela and Tamrat Bekele, 2002; Leul Woldemichael *et al.*, 2010; Fitsum Temesgen *et al.*, 2021). Then, the disturbance scores for each forest patch were assigned to reflect the highest rate of disturbance and the absence of disturbance.

Vegetation structure analysis: The structural analysis of vegetation was described using species density, DBH, height, basal area (BA) per hectare, frequency, and important value index (IVI), following Tamrat Bekele (1993). The frequency distribution of a species was computed as the proportion of samples within which a species is found. The dominance of key woody species was measured using the Importance Value Index (IVI), considering three factors: relative density, relative dominance, and relative frequency (Kent and Coker, 1992).

Different formulas that are important in conducting structural analysis are described below:

$$\boldsymbol{Density\ (D)} = \frac{The\ number\ of\ individuals\ of\ species}{Unit\ area\ of\ the\ quadrat\ (hectare)}$$

$$\boldsymbol{Relative\ density\ (RD)} = \frac{Number\ of\ individuals\ of\ species\ A}{Total\ number\ of\ individuals\ of\ all\ species} \times 100$$

$$\boldsymbol{Frequency\ (F)} = \frac{Number\ of\ sampling\ units\ or\ quadrats\ in\ which\ species\ A\ recorded}{Total\ number\ of\ sampling\ units\ or\ quadrats\ sampled}$$

$$\boldsymbol{Relative\ frequency\ (RF)} = \frac{frequency\ of\ single\ species}{Sum\ of\ frequencies\ of\ all\ species} \times 100$$

$$\boldsymbol{Basal\ Area\ (BA)} = \frac{\pi d^2}{4}$$; Where, $\pi = 3.14$; d = DBH (m)

$$\boldsymbol{Dominance\ (Do)} = \frac{Area\ covered\ by\ a\ species\ (BA)}{Sum\ of\ all\ quadrats\ area\ in\ hectare}$$

$$\boldsymbol{Relative\ dominance\ (RDo)} = \frac{Basal\ area\ of\ a\ single\ species}{Total\ basal\ area\ of\ all\ species} \times 100$$

$$\boldsymbol{Importance\ Value\ Index\ (IVI)} = RD\ +\ RDo +\ RF$$

Where, IVI = Importance Value Index; RD = Relative Density; RDo = Relative Dominance and RF = Relative Frequency

3.3. Results

3.3.1. Composition of woody species in the forest patches

A total of 55 woody plant species belonging to 31 families and 46 genera were recorded in the study area (Table 2.1 and Table 2.2). The highest species richness was recorded for the Fabaceae family 12 (21.8%), followed by the Anacardiaceae, Euphorbiaceae, Malvaceae, and Moraceae with 3 (5.5%) each (Figure 2.2). Among the total woody species recorded in the study areas, 28 (51%) were trees, and 27 (49%) were found to be shrubs. The highest richness among the forest patches was recorded in Gerado forest, followed by Micha and Mekelet forest patches. Gerado has 50 woody plant species belonging to 44 genera and 32 families. Micha has 32 woody plant species belonging to 27 genera and 19 families. Mekelet also has 21 plant species belonging to 20 genera and 16 families.

Table 2.1: Total number of species, genera, families, and growth forms per forest patch

Patch name	Area (ha)	Altitudinal range (masl)	No. of plots	Number of transects	Richness	Family	Genera	Growth form	
								Tree	Shrub
Gerado	75	1925 - 2226	43	12	50	32	44	24	26
Micha	44.2	1997 - 2226	28	9	32	19	27	14	18
Mekelet	92	2059 - 2413	24	5	21	16	20	9	12

Table 2.2: List of woody plant species recorded from Gerado, Micha and Mekelet forest patches, North Wollo Zone, Ethiopia

Species name	Family	Local name	Growth form (GF)	Occurrence in the forest patches
Acacia decurrens (J.C.Wendl.) Willd.	Fabaceae	Akacha	T	1
Afrocarpus falcatus (Thunb.) C.N.Page	Podocarpaceae	Zigba	T	1
Albizia gummifera (J.F.Gmel.) C.A.Sm.	Fabaceae	Sesa	T	1
Allophylus abyssinicus (Hochst.) Radlk.	Sapindaceae	Embis	T	1, 2, 3
Arundo donax L.	Poaceae	Shenbeko	S	1
Calotropis procera (Aiton) W.T.Aiton	Apocynaceae	Tobia	S	1
Calpurnia aurea (Aiton) Benth.	Fabaceae	Digita	S	1
Carissa spinarum L.	Apocynaceae	Agam	S	1, 2, 3
Casuarina equisetifolia L.	Casuarinaceae	Shiwshiwa	T	1
Celtis africana Burm.f.	Cannabaceae	Awrarisie	S	1
Citrus aurantiifolia (Christm.) Swingle	Rutaceae	Lomi	S	1
Cordia africana Lam.	Boraginaceae	Wanza	T	1

Species name	Family	Local name	Growth form (GF)	Occurrence in the forest patches
Croton macrostachyus Hochst. ex Delile	Euphorbiaceae	Mekanisa	T	2, 3
Dichrostachys cinerea (L.) Wight & Arn.	Fabaceae	Gorgoro/Ader	S	2
Dodonaea viscosa subsp. angustifolia (L.f.) J.G.West	Sapindaceae	Kitkita	S	1, 2, 3
Dombeya torrida (J.F.Gmel.) Bamps	Malvaceae	Danisa/Kuncho	S	1,2
Ehretia cymosa Thonn	Boraginaceae	Ulaga	T	1
Eucalyptus camaldulensis Dehnh.	Myrtaceae	Key Bahirzaf	T	1, 2, 3
Euclea racemosa L.	Ebenaceae	Dedeho	S	1, 2, 3
Euphorbia abyssinica J.F.Gmel.	Euphorbiaceae	Qulqual	S	1, 2, 3
Euphorbia tirucalli L.	Euphorbiaceae	Kinchibt	S	1, 2
Faidherbia albida (Delile) A.Chev.	Fabaceae	Gerbi	T	1, 3
Ficus capreifolia Delile	Moraceae	Beles	T	2
Ficus sur Forssk.	Moraceae	Shola	T	2
Ficus vasta Forssk.	Moraceae	Warka	T	1
Galiniera saxifraga (Hochst.) Bridson	Rubiaceae	Buna Mesay	S	1
Grevillea robusta A.Cunn. ex R.Br.	Proteaceae	Gravilea	T	1
Grewia bicolor Juss.	Malvaceae	Sefa	S	1
Grewia ferruginea Hochst. ex A.Rich.	Malvaceae	Lenquata	S	1, 2, 3
Gymnanthemum amygdalinum (Delile) Sch.Bip.	Asteraceae	Girawa	S	1, 2, 3
Gymnosporia senegalensis (Lam.) Loes.	Celastraceae	Atat	S	1, 2, 3
Hesperocyparis lusitanica (Mill.) Bartel	Cupressaceae	Yeferenj Tsid	T	1, 2

Species name	Family	Local name	Growth form (GF)	Occurrence in the forest patches
Heteromorpha arborescens (Spreng.) Cham. & Schltdl.	Apiaceae	Yejib Mirkuz	S	1, 2
Jacaranda mimosifolia D.Don	Bignoniaceae	Yetebenja Zaf /Jakaranda	T	1
Juniperus procera Hochst. ex Endl.	Cupressaceae	Yehabesha Tsid	T	1, 2, 3
Moringa oleifera Lam.	Moringaceae	Shiferaw/Moringa	T	1
Myrsine africana L.	Primulaceae	Kechem	S	1, 2, 3
Olea europaea L. subsp. *cuspidata* (Wall. & G.Don) Cif.	Oleaceae	Woyra	T	1, 2, 3
Opuntia ficus-indica (L.) Mill.	Cactaceae	Qulqual / Manka/Beles/Wedi	S	1
Osyris lanceolata Hochst. & Steud.	Santalaceae	Keret	S	1, 2, 3
Pittosporum viridiflorum Sims	Pittosporaceae	Kefeto	T	1, 2, 3
Premna schimperi Engl.	Lamiaceae	Checho	S	1, 3
Pterolobium stellatum (Forssk.) Brenan	Fabaceae	Kentafa	S	1, 2, 3
Rhamnus prinoides L'Hér.	Rhamnaceae	Gesho	S	1
Rosa abyssinica R.Br. ex Lindl.	Rosaceae	Kega	S	1, 2
Schinus molle L.	Anacardiaceae	Kundo-berberie	T	1, 2
Searsia glutinosa (Hochst. ex A.Rich.) Moffett	Anacardiaceae	Embis/2	S	1, 2
Searsia retinorrhoea (Steud. ex Oliv.) Moffett	Anacardiaceae	Talo	S	1, 2, 3
Senegalia brevispica (Harms) Seigler & Ebinger	Fabaceae	Kontr	S	1, 2

Species name	Family	Local name	Growth form (GF)	Occurrence in the forest patches
Vachellia amythethophylla (Steud. ex A.Rich.) Kyal. & Boatwr.	Fabaceae	Lafto	T	1
Vachellia etbaica (Schweinf.) Kyal. & Boatwr.	Fabaceae	Tikur Girar	T	1, 2, 3
Vachellia seyal (Delile) P.J.H.Hurter	Fabaceae	Wacho Girar	T	2
Vachellia sieberiana (DC.) Kyal. & Boatwr.	Fabaceae	Key Girar	T	1, 2, 3
Vachellia tortilis (Forssk.) Galasso & Banfi	Fabaceae	Deweni Grar, Korera	T	1
Ziziphus spina-christi (L.) Desf.	Rhamnaceae	Kurkura, Geba	T	1, 2

Key: Growth form, GF (Tree, T; Shrub, S). Forest patch, FP (1, Gerado forest patch; 2, Micha forest patch; 3, Mekelet forest patch)

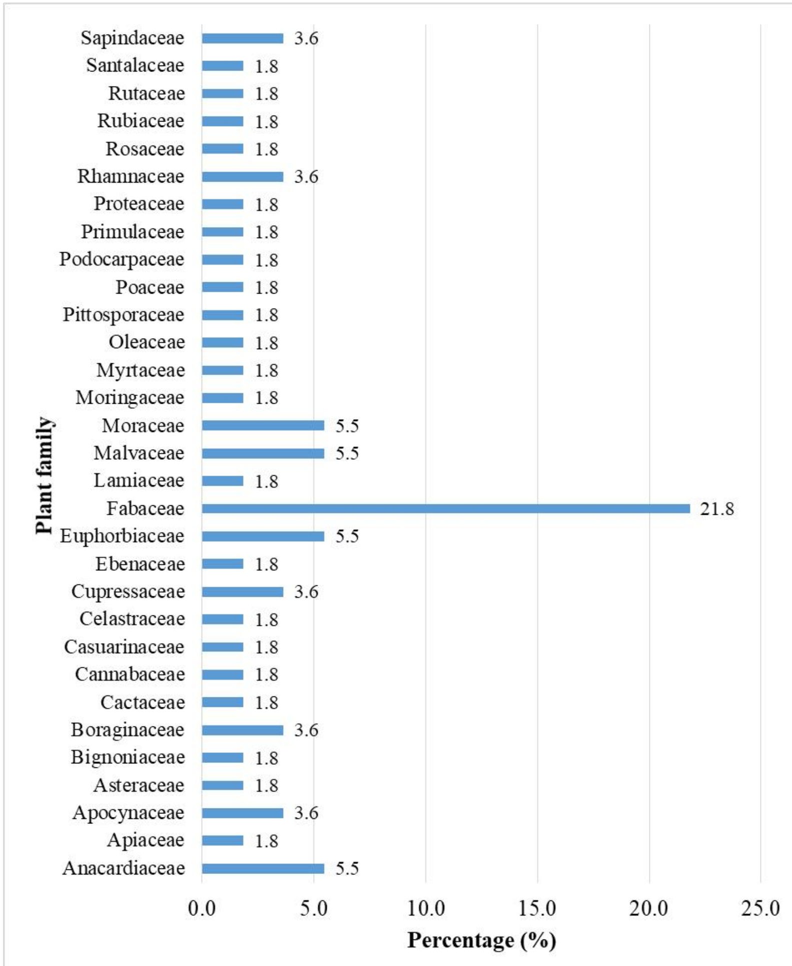

Figure 2.2: Species richness of plant family in percentage (%)

3.3.2. Plant communities of the forest patches

In this study, four plant community types were identified (Figure 3). There is a statistically significant difference ($R = 0.4703,\ P \leq 0.001$) in floristic compositions of the woody species among the four identified plant community types in all forest patches based on the Multi-response Permutation Procedures (MRPP) test. Each plant community types (C1-C4) were named after the two identifying characteristics of species in each group (Table 3). The detailed descriptions of the community types identified from Gerado, Micha and Mekelet forest patches are given below.

(i): *Hesperocyparis lusitanica - Juniperus procera* community type (C1): This community type represents 13 plots and 25 species found between the altitudinal ranges of 1925 – 2204 m.a.s.l. The dominant tree and shrub species of this community type include *Juniperus procera, Vachellia sieberiana, Dodonaea viscosa subsp. angustifolia, Olea europaea L. subsp. cuspidata, Hesperocyparis lusitanica, Eucalyptus camaldulensis, Euclea racemosa, Pittosporum viridiflorum and Osyris lanceolata.*

(ii): *Pittosporum viridiflorum - Dodonaea viscosa subsp. angustifolia* community Type (C2): This community type represents 19 plots and 28 species between the altitudinal ranges of 1929 - 2178 m.a.s.l. The dominant tree and shrub species of this community type include *Vachellia sieberiana, Pittosporum viridiflorum*, *Dodonaea viscosa* subsp. *angustifolia, Searsia retinorrhoea, Olea europaea* L. subsp. *cuspidata*, *Carissa spinarum, Vachellia etbaica* and *Euclea racemosa.*

(iii): *Olea europaea L. subsp. cuspidata - Allophylus abyssinicus* community type (C3): This community type represents 12 plots and 45 species between the altitudinal ranges of 1946 – 2226 m.a.s.l. The dominant tree and shrub species of this community type include *Olea europaea* L. *subsp. cuspidata, Dodonaea viscosa* subsp. *angustifolia, Vachellia sieberiana, Pittosporum viridiflorum, Allophylus abyssinicus, Juniperus procera, Carissa spinarum, Euphorbia abyssinica* and *Euclea racemosa.*

(iv): *Vachellia sieberiana - Carissa spinarum* community type (C4): This community type represents 51 plots and 31 species between the altitudinal ranges of 1937 – 2413 m.a.s.l. The dominant tree and shrub species of this community type include *Vachellia sieberiana, Dodonaea viscosa* subsp. *angustifolia, Olea europaea* L. subsp. *cuspidata, Carissa spinarum, Eucalyptus camaldulensis, and Osyris lanceolata.*

3.3.3. Plant community relationship with environmental variables

In this study, Redundancy Analysis (RDA) was utilized to examine the relationships between community types and environmental variables. The RDA results showed that slope, altitude and cutting had significant impacts ($p \leq 0.05$) on the composition and distribution of community types (Figure 2.4; Table 2.5). The RDA result showed that the community type 4 was primarily correlated with cutting, whereas the community type 2 was primarily correlated with altitude and slope (Figure 2.4).

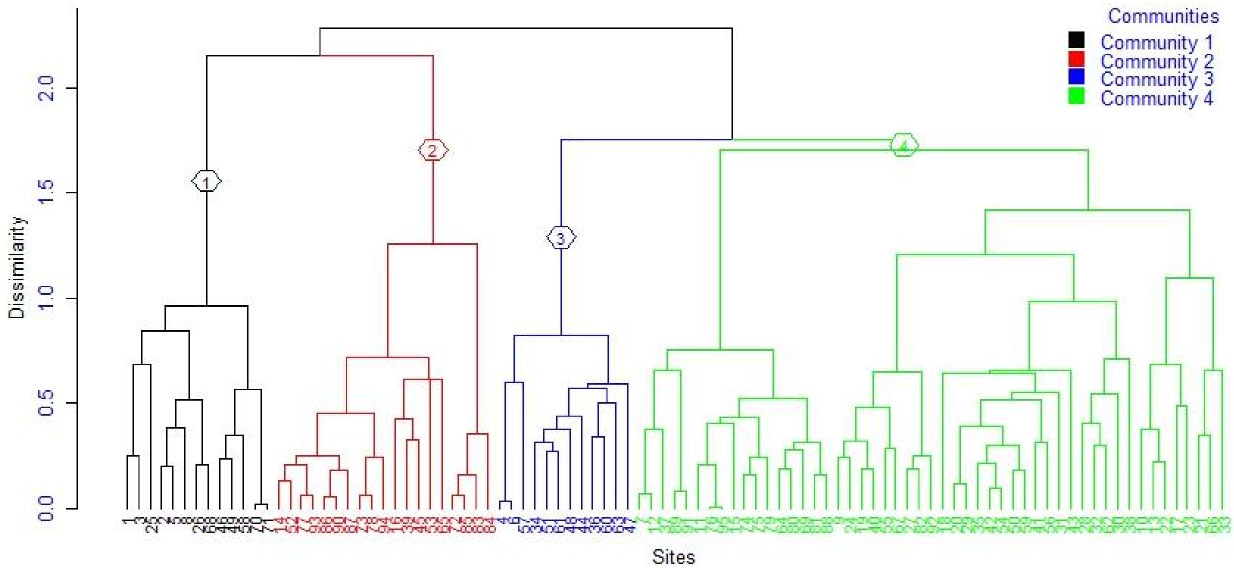

Figure 2.3: Dendrogram showing the four plant community types of the forest patches

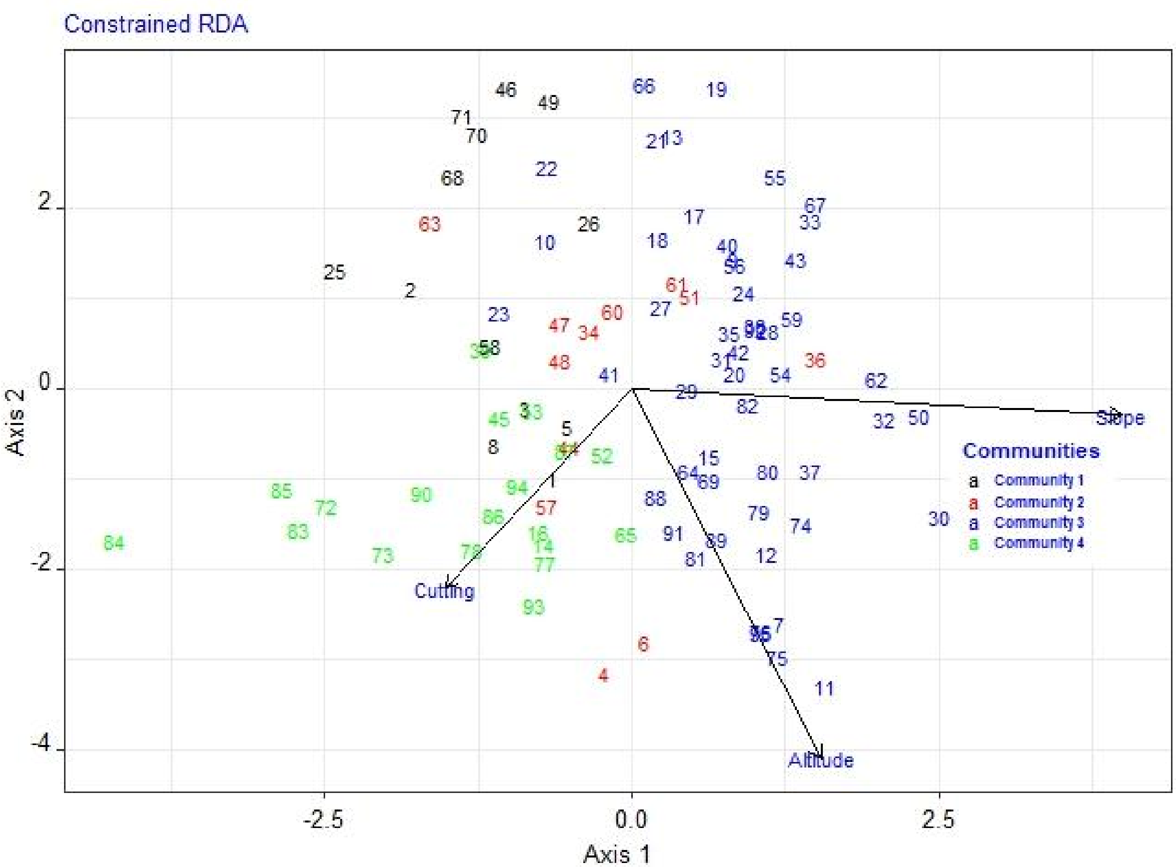

Figure 2.4: RDA of community types with significant environmental variables (p ≤0.05)

Table 2.3: Characteristic species in each community type of forest patches based on the average synoptic cover-abundance value

Characteristics species	**Community types**			
	C1	**C2**	**C3**	**C4**
Hesperocyparis lusitanica	**1.92**	0.07	0.04	0.53
Juniperus procera	**7**	0.67	1.38	0.21
Pittosporum viridiflorum	1.62	**5.07**	2.9	0.53
Dodonaea viscosa subsp. *angustifolia*	2.92	**4.93**	4.21	3.47
Olea europaea L. subsp. *cuspidata*	2.54	2.13	**5.52**	3.16
Allophylus abyssinicus	0.77	0.27	**1.4**	0.21
Vachellia sieberiana	4.08	5.4	3.15	**5.63**
Carissa spinarum	0	1.2	1.0	**0.68**

Table 2.4: Detrended correspondence analysis (DCA) result of vegetation data

	DCA1	DCA2	DCA3	DCA4
Eigenvalues	0.2528	0.2150	0.1750	0.1815
Decorana values	0.3167	0.2421	0.1914	0.1533
Axis lengths	2.7942	2.8080	2.3104	2.0698

Table 2.5: Outcomes of the Adonis 2 test on significant environmental variables

```
          Df SumsOfSqs MeanSqs F.Model      R2 Pr(>F)    
Slope      1    1.3412 1.34116  9.2263 0.08536  0.001 ***
Aspect     1    0.2709 0.27090  1.8636 0.01724  0.053 .  
Cutting    1    0.4815 0.48151  3.3125 0.03065  0.001 ***
Grazing    1    0.1468 0.14679  1.0098 0.00934  0.420    
Browsing   1    0.1483 0.14830  1.0202 0.00944  0.413    
Altitude   1    0.5818 0.58175  4.0021 0.03703  0.001 ***
FWC        1    0.2552 0.25519  1.7556 0.01624  0.068 .  
CP         1    0.0457 0.04571  0.3145 0.00291  0.969    
AE         1    0.0844 0.08438  0.5805 0.00537  0.799    
Residuals 85   12.3559 0.14536         0.78642           
Total     94   15.7116                 1.00000           

Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

3.3.4. Floristic diversity indices

The results reveal that Gerado has the highest species richness, followed by Micha and then Mekelet. Among the forest patches, the highest diversity was observed in community type 3 (2.752), followed by community type 2 (2.56). The Shannon evenness value was also in the same order with little variations, the highest being for community type 1 (0.779) and community type 2 (0.777) and the least evenness was for community type 4 (Table 6).

Table 2.6: Species richness and diversity indices of plant communities identified in the forest patches in the study area

Forest Patch	Communities	Richness	H	Shannon Evenness	Simpson Evenness
Gerado, Micha and Mekelet (All in one)	C1	25	2.506	0.779	0.349
	C2	27	2.560	0.777	0.319
	C3	45	2.752	0.723	0.226
	C4	31	2.471	0.720	0.240

3.3.5. Floristic composition similarities between forest patches

The similarities between forest patches were generally strong. Accordingly, Gerado and Micha patch forests exhibited the highest similarity (Table 2.7 and Figure 2.5). Moreover, the Micha patch forest showed strong similarities with Mekelet (Ss = 0.71). Relatively the least similarity was observed between Gerado and Mekelet patch forests.

Table 2.7: Floristic composition similarity indices of the three forest patches

Forest patches	Gerado	Micha	Mekelet
Gerado	***	0.6585	0.5634
Micha		***	0.7170
Mekelet			***

*** indicates 1.0000 (100%) similarity.

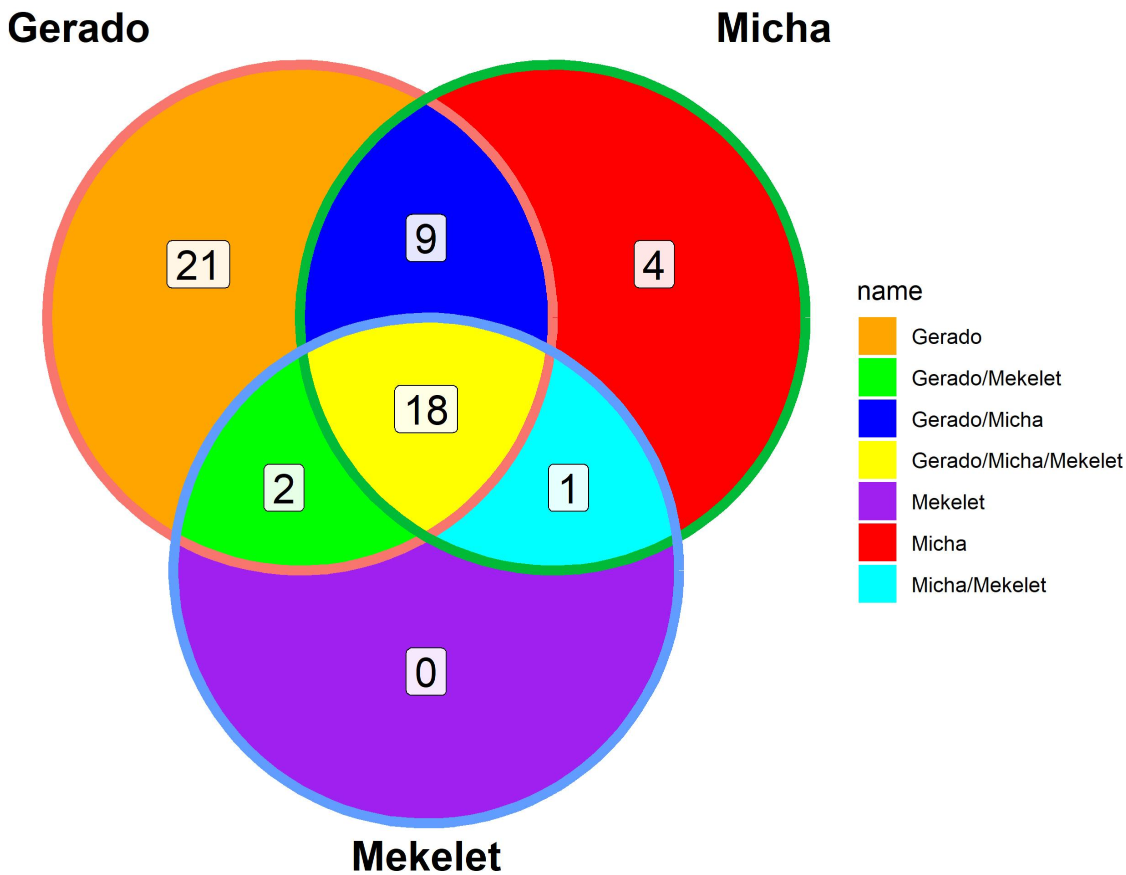

Figure 2.5: Venn-Diagram of species in Gerado, Micha and Mekelet forest patches

3.3.6. Anthropogenic disturbances in the forest patches

The anthropogenic disturbance levels in the three forest patches varied from a maximum mean score total of 3.96 for Mekelet and a minimum score of 2.02 for Micha and 2.96 for Gerado Forest patch (Table 2.8). The Mekelet forest patch shows the highest mean disturbance score at 0.66, indicating a moderate to high overall impact. The intensity of cutting (mean score 1.71) and firewood collection (mean score 0.75) significantly contribute to the elevated disturbance level in this patch. Micha forest patch exhibits a mean disturbance score of 0.34, suggesting a relatively lower overall impact compared to the Gerado. Cutting (mean score 0.67) and agricultural expansion (mean score 0.56) are the primary contributors, indicating a moderate level of disturbance. In the Gerado forest patch, the mean disturbance score is 0.49, reflecting a relatively low to moderate overall impact. This result is influenced by notable contributions from cutting and agricultural expansion, indicating human activities are moderately affecting this patch.

Table 2.8: Mean value of anthropogenic disturbances along the three forest patches

Patch name and total number of plots	Reported plots and score value (mean)	Cutting	Charcoal production	Agricultural expansion	Firewood collection	Grazing	Browsing	Total
Gerado, 43 plots	No. of plots	25	0	13	14	6	9	
	Mean	1	0.04	0.82	0.46	0.39	0.25	2.96
Micha, 28 plots	No. of plots	19	1	13	8	11	7	
	Mean	0.67	0.00	0.56	0.35	0.19	0.26	2.02
Mekelet, 24 plots	No. of plots	22	5	9	11	3	4	
	Mean	1.71	0.38	0.67	0.75	0.17	0.29	3.96

3.3.7. Vegetation structure

Density

In the Gerado forest patch, the most abundant species is *Dodonaea viscosa* subsp. *angustifolia*, followed by *Olea europaea* L. subsp. *cuspidata*. The species with the highest RD is also *Dodonaea viscosa* subsp. *angustifolia*, indicating its significant presence and ecological influence within the Gerado patch. Micha's species distribution is similar to Gerado's, with *Dodonaea viscosa* subsp. *angustifolia* and *Olea europaea* L. subsp. *cuspidata* again being the most abundant. However, *Dodonaea viscosa* subsp. *angustifolia* has the highest RD here, which could suggest it has a more significant impact on the ecosystem's structure in Micha. Mekelet shows a different pattern, where *Vachellia sieberiana* has the highest number of individuals and RD, indicating its dominance in this patch (Table 2.9).

Table 2.9: Density and relative density of top 5 woody plant species across the three forest patches

Patch name	Species name	No. of individuals	D	Total No. of stems	RD	Rank
Gerado	*Dodonaea viscosa* subsp. *angustifolia*	893	519.19	3957	22.6	1
	Olea europaea L. subsp. *cuspidata*	679	394.77		17.2	2
	Pittosporum viridiflorum	473	275.00		12.0	3
	Vachellia sieberiana	263	152.91		6.6	4
	Euclea racemosa	243	141.28		6.1	5
Micha	*Dodonaea viscosa* subsp. *angustifolia*	614	548.21	2158	28.5	1
	Olea europaea L. subsp. *cuspidata*	243	216.96		11.3	2

	Vachellia sieberiana	152	135.71		7.0	3
	Pittosporum viridiflorum	136	121.43		6.3	4
	Juniperus procera	132	117.86		6.1	5
Mekelet	*Dodonaea viscosa* subsp. *angustifolia*	800	833.333	2023	39.5	1
	Vachellia sieberiana	441	459.375		21.8	2
	Olea europaea L. subsp. *cuspidata*	292	304.167		14.4	3
	Eucalyptus camaldulensis	136	141.667		6.7	4
	Osyris lanceolata	57	59.375		2.8	5

Frequency

The Gerado and Micha share a similar species hierarchy, with these species occurring in over 85% of plots, reflective of their adaptability and potential influence on local biodiversity. *Dodonaea viscosa* subsp. *angustifolia* and *Vachellia sieberiana* are the most frequent species, both occurring in 95.35% of plots, with the highest RF of 10.17, indicating their widespread presence and possible dominance in the forest structure in Gerado. Other species like *Pittosporum viridiflorum*, *Olea europaea* L. subsp. *cuspidata*, and *Euclea racemosa* L. also show high frequency, suggesting a diverse but specific set of dominant species in this patch.

The same two species, *Dodonaea viscosa* subsp. *angustifolia* and *Vachellia sieberiana*, also show the highest frequency and RF in Micha, pointing towards a similar species dominance as in Gerado. However, the presence of *Allophylus abyssinicus* with a lower frequency and RF indicates a variation in the understory composition between Micha and Gerado. Similarly, in Mekelet, *Vachellia sieberiana* shows the highest frequency and RF, present in 100% of the plots (Table 2.10).

Table 2.10: Frequency and relative frequency of top 5 woody plant species across the three forest patches

Patch name	**Species name**	**No. of individuals**	**No of plots species occurs**	**Frequency (%)**	**∑ of F of all spp.**	**RF**	**Rank**
Gerado	*Dodonaea viscosa* subsp. *angustifolia*	893	41	95.35	937.209	10.17	1
	Vachellia sieberiana	263	41	95.35		10.17	1
	Pittosporum viridiflorum	473	39	90.70		9.68	3
	Olea europaea L. subsp.	679	36	83.72		8.93	4

	cuspidata						
	Euclea racemosa	243	34	79.07		8.44	5
	Dodonaea viscosa subsp. *angustifolia*	614	24	85.71		10.08	1
	Vachellia sieberiana	152	24	85.71		10.08	1
Micha	*Olea europaea* L. subsp. *cuspidata*	243	23	82.14	850	9.66	3
	Pittosporum viridiflorum Sims	136	22	78.57		9.24	4
	Allophylus abyssinicus	95	16	57.14		6.72	5
	Vachellia sieberiana	441	24	100		15.09	1
	Dodonaea viscosa subsp. *angustifolia*	800	22	91.67		13.84	2
Mekelet	*Olea europaea* L. subsp. *cuspidata*	292	20	83.33	662.5	12.58	3
	Allophylus abyssinicus	46	13	54.17		8.18	4
	Eucalyptus camaldulensis	136	12	50		7.55	5

Basal area

The total basal area (BA m^2) of the forest patches was also determined from the DBH values (cm ha^{-1}). The total basal area of Gerado, Micha and Mekelet forest patches calculated was found to be 25.976 m^2 ha^{-1}, 13.549 m^2 ha^{-1} and 15.45 m^2 ha^{-1} respectively. The BA was determined for individual woody species in Micha forest patch varied from 2.84 to 0.02 m^2 ha^{-1} and Mekelet from 7.25 to 0.01 m^2 ha^{-1} and Gerado 5.41 to 0.02 m^2 ha^{-1} (Table 2.11).

In the Gerado forest patch, *Olea europaea* L. subsp. *cuspidata* stands out as the most structurally influential species with the highest basal area (BA) and dominance, followed by *Pittosporum viridiflorum* and *Vachellia sieberiana* which also play significant roles in shaping the forest's structure, whereas *Dodonaea viscosa* subsp. *angustifolia*, despite having the lowest BA among the top five, maintains a notable influence. In contrast, Micha is characterized by the dominance of *Vachellia sieberiana*, with *Juniperus procera* and *Olea europaea* L. subsp. *cuspidata* also features prominently, and *Euphorbia abyssinica* appears as the least dominant among the top species, suggesting a diversity in species roles within the ecosystem. Similarly, Mekelet's vegetation structure is governed by the dominance of *Vachellia sieberiana*, with *Olea europaea* L. subsp. *cuspidata* and *Dodonaea viscosa* subsp. *angustifolia* following in BA, and *Juniperus procera* occupying a smaller yet significant ecological niche as one of the top five species.

Table 2.11: Basal Area (BA) and dominance of top five woody plant species in Gerado, Micha and Mekelet forest patches

Patch name	Scientific Name	T BA of spp	BA (m^2h^{-1})	Dominance	∑ of all BA	∑ of all BA	RDO	Rank
Gerado	*Olea europaea* L. subsp. *cuspidata*	54174.318	5.417	31496.70	259763.73	25.98	20.855	1
	Pittosporum viridiflorum	49564.500	4.956	28816.57			19.081	2
	Vachellia sieberiana	48566.568	4.857	28236.38			18.696	3
	Juniperus procera	43025.159	4.303	25014.63			16.563	4
	Dodonaea viscosa subsp. *angustifolia*	12754.060	1.275	7415.15			4.910	5
Micha	*Vachellia sieberiana*	28418.04	2.842	25373.25	135487.61	13.55	20.97	1
	Juniperus procera	22534.79	2.253	20120.35			16.63	2
	Olea europaea L. subsp. *cuspidata*	20820.55	2.082	18589.77			15.37	3
	Eucalyptus camaldulensis	18505.01	1.851	16522.33			13.66	4
	Euphorbia abyssinica	10482.23	1.048	9359.13			7.74	5
Mekelet	*Vachellia sieberiana*	72534.16	7.25	75556.42	154505.47	15.45	46.95	1
	Olea europaea L. subsp. *cuspidata*	35726.86	3.57	37215.48			23.12	2
	Dodonaea viscosa subsp. *angustifolia*	17908.01	1.79	18654.18			11.59	3
	Eucalyptus camaldulensis	12138.45	1.21	12644.22			7.86	4
	Juniperus procera	4363.27	0.44	4545.07			2.82	5

Diameter at breast height (DBH) structure of the forest patches

In the smallest DBH class (2.5 - 9.0 cm), representing younger or smaller trees, all three sites showed a high percentage of their tree populations (Figure 2.6). Micha (71.9%) had the highest proportion of trees in this class, followed closely by Gerado (70.3%) and then Mekelet (62.6%). This indicates a strong presence of younger trees across all sites, with Micha having a slightly more significant proportion of these younger trees. The slightly higher percentage in Micha could indicate either a more diverse range of young species or a more conducive environment for young tree growth.

As the tree size increases, there is a notable decrease in the percentage of trees in all sites. However, Mekelet stands out with a relatively higher percentage (31.9%) in the 9.1 - 17.0 cm class compared to Gerado (23.8%) and Micha (23.3%). This could imply that Mekelet has a more substantial intermediate-sized tree population. The trend continues in the 17.1 - 25.0 cm class, though the differences among the sites become less pronounced. In contrast, the lower percentages in Gerado and Micha could imply either a less diverse range of intermediate-sized species or factors limiting their growth.

In the larger DBH classes (25.1 cm – 43.0 cm), which indicate mature, large-diameter trees, all sites show minimal percentages. Gerado, Micha, and Mekelet exhibit a similar pattern, suggesting a limited presence of older, larger trees across the sites. This could suggest limited species diversity in the mature tree segment, possibly due to historical logging, anthropogenic factors (cutting, charcoal production, firewood collection), or other ecological factors. The similar patterns across the sites might reflect a regional trend or a shared ecological history.

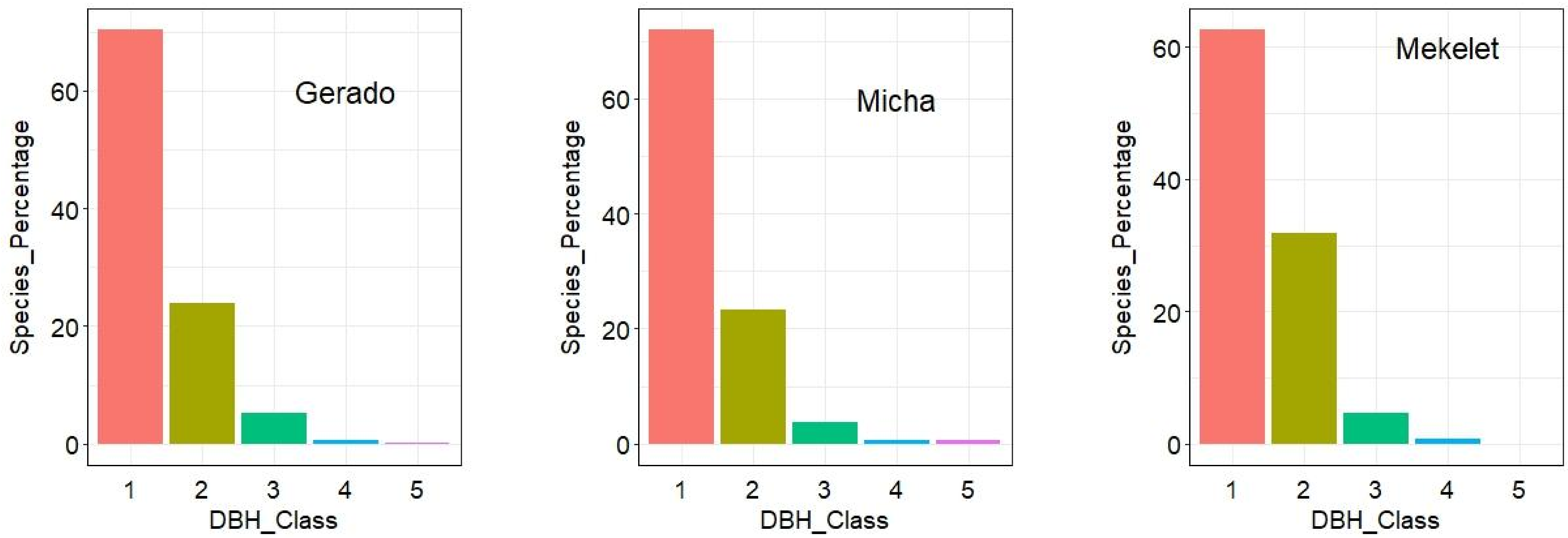

Figure 2.6: Distribution of woody species density among DBH classes (1 = 2.5 - 9.0 cm, 2 = 9.1-17.0 cm, 3 = 17.1 – 25.0 cm, 4 = 25.1- 32 cm, 5 = 32.1 – 43.0 cm)

Height class of the species in the study site

Across the forest patches of Gerado, Micha, and Mekelet, a pronounced dominance of shorter trees (2.0 - 6.0 meters) was observed, with respective percentages of 85.82%, 90.47%, and 90.06% (Figure 2.7). This suggests a youthful or regenerating forest demographic, potentially influenced by a combination of environmental conditions, forest management practices by the local community, or recent growth patterns. While the specific management strategies, if any, employed within these forests remain unclear, it is plausible that practices such as selective logging, controlled burns, or reforestation efforts could contribute to the observed patterns. The subsequent height class (6.1 - 10.0 meters) shows a substantial decline in tree population, with Gerado at 12.51%, Micha at 8.64%, and Mekelet at 9.29%, reflecting a common ecological trend where fewer trees reach mid-sized maturity.

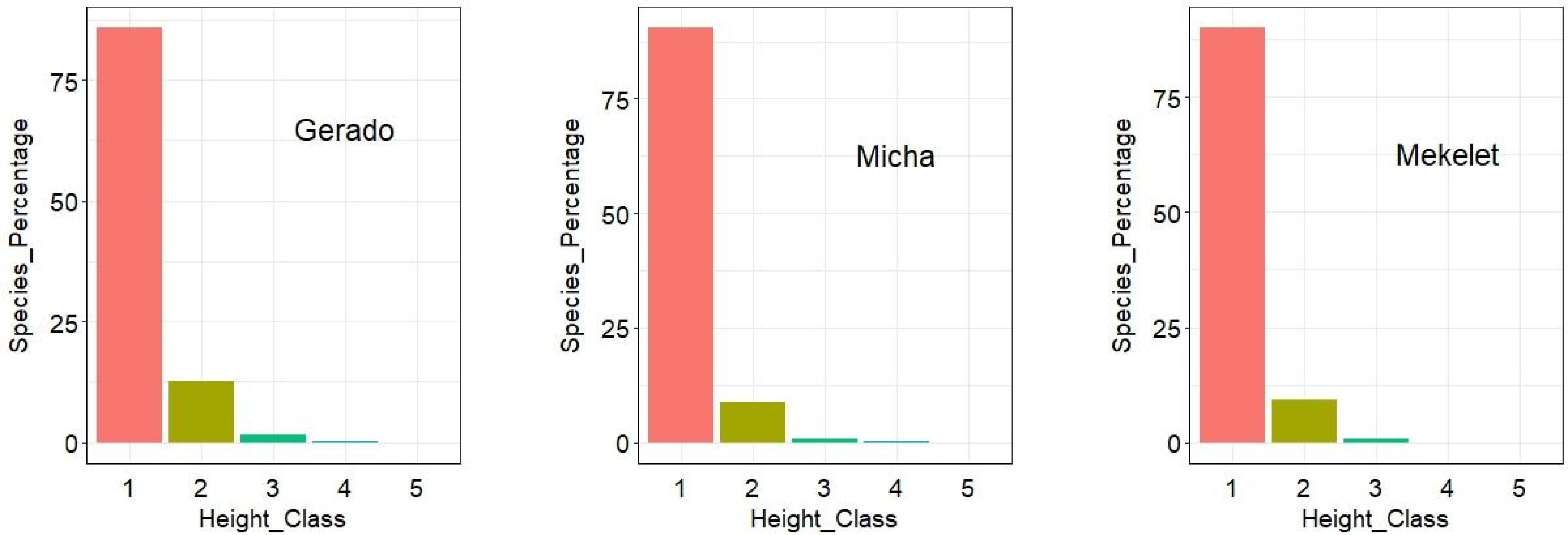

Figure 2.7: Frequency distribution by height class of woody species (1 = 2.0 - 6.0 m, 2 = 6.1-10.0 m, 3 = 10.1 – 15.0 m, 4 = 15.1- 20.0 m, 5 = 21.1 – 26.0 m)

Importance value index (IVI)

The most important species in Gerado is *Olea europaea* L. *subsp. cuspidata*, showing the highest IVI of 46.95. This indicates its dominance and prevalence in terms of density, frequency, and canopy cover within the Gerado patch. Other notable species include *Pittosporum viridiflorum* and *Dodonaea viscosa* subsp. *angustifolia*, which also holds significant importance within this patch. The variety among the top species suggests a diversity of trees contributing to the ecological makeup of Gerado. In Micha, *Dodonaea viscosa* subsp. *angustifolia* ranks as the most important species, with an IVI of 43.23 (Table 2.12). Like Gerado, *Vachellia sieberiana* and *Olea europaea* L. subsp. *cuspidata* are also important, although their IVIs are rearranged.

Table 2.12: Top five IVI value-ranked species in each forest patches

Patch name	Species name	RD	RF	RDO	IVI	Rank
Gerado	*Olea europaea* L. subsp. *cuspidata*	17.16	8.93	20.86	46.95	1
	Pittosporum viridiflorum	11.95	9.68	19.08	40.71	2
	Dodonaea viscosa subsp. *angustifolia*	22.57	10.17	4.91	37.65	3
	Vachellia sieberiana	6.65	10.17	18.70	35.52	4
	Juniperus procera	4.04	2.98	16.56	23.58	5
Micha	*Dodonaea viscosa* subsp. angustifolia	27.83	10.08	5.31	43.23	1
	Vachellia sieberiana	6.89	10.08	20.97	37.95	2
	Olea europaea L. subsp. *cuspidata*	11.02	9.66	15.37	36.05	3
	Juniperus procera	5.98	6.30	16.63	28.92	4
	Eucalyptus camaldulensis	4.71	5.46	13.66	23.83	5
Mekelet	*Vachellia sieberiana*	21.80	15.09	46.95	83.84	1
	Dodonaea viscosa subsp. *angustifolia*	39.55	13.84	11.59	64.97	2
	Olea europaea L. subsp. *cuspidata*	14.43	12.58	23.12	50.14	3
	Eucalyptus camaldulensis	6.72	7.55	7.86	22.13	4
	Allophylus abyssinicus	2.27	8.18	0.75	11.20	5

Key: Relative density, RD; Relative dominance, RDO; Relative frequency, RF; Important value index, IVI

Population structure of some selected woody plant species

In this study we identified three population structure patterns based on the distribution of diameter at breast height (DBH) classes in selected species (Figure 2.8). These patterns include the inverted J-shaped pattern, broken inverted J-shaped pattern and bell-shaped pattern. The inverted J-shaped pattern exhibited the highest frequencies in lower size classes and the gradual declining towards higher size classes. Examples of species displaying this pattern include *Olea europaea* L. *subsp. cuspidata and Pittosporum viridiflorum.* Broken inverted J-shaped pattern showed the percentage of individuals in the lower DBH class 1 and 2 is very high but becoming lower in the highest DBH classes even nothing in some DBH class 3, 4 and 5. This pattern is manifested by *Dodonaea viscosa* subsp. *angustifolia*. The bell-shaped pattern also showed species where the highest percentages of individuals are in the middle DBH class and smaller percentages in the lower and higher DBH classes. This pattern is manifested by *Vachellia sieberiana*.

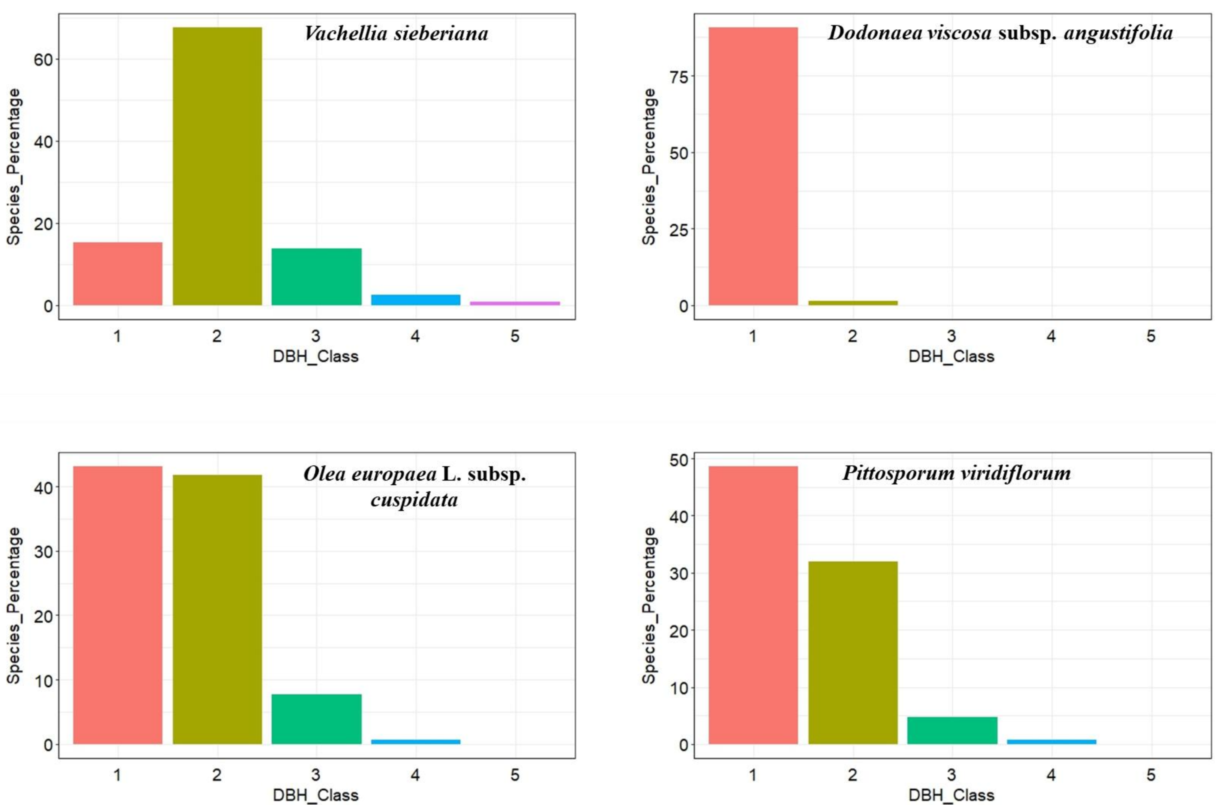

Figure 2.8: Population structure pattern of selected woody plant species in the study area

3.4. Discussion

3.4.1. Floristic composition

Our findings suggest that the woody species richness in the study area is lower (55 woody plant species) compared to other similar studies conducted on the dry Afromontane forests of Ethiopia, e.g. Dindin Natural Forest, southeast of Ethiopia (Tamiru Lemi *et al.*, 2023), Melit, Addisinagulit, and Yetegan (Megabzer) forest patches, Gozamin district of Northwest Ethiopia (Metsehet Yinebeb *et al.*, 2023), Guard dry Afromontane forest of northwestern Ethiopia (Yitayih Dagne and Liyew Birhanu, 2023), Gennemar dry Afromontane forest in southern Ethiopia (Shemsu Ahmed *et al.*, 2022), Kafta Sheraro National Park Dry Forest, Tigray Region, Ethiopia (Fitsum Temesgen *et al.*, 2021), Gelawoldie community forest, northwestern Ethiopia (Getie Mucheye and Getahun Yemata, 2020), dry Afromontane forests of Awi Zone, northwestern Ethiopia (Getaneh Gebeyehu *et al.*, 2019), Wof Washa natural forest, northeast Ethiopia (Gebremicael Fisaha *et al.*, 2013). These studies have examined various aspects of the forests, including plant community structure, woody species composition, environmental determinants, tree growth, and regeneration status of tree species. The variations in species richness among these forests could be due to diverse factors, including altitudinal range, geographical location, climate, topography, environmental heterogeneity, regeneration success, competition, anthropogenic disturbances, and edaphic influences (Schmitt *et al.*, 2013; Metsehet Yinebeb *et al.*, 2023; Zeleke Assefa *et al.*, 2023). The plant family Fabaceae, which turned out to be the most dominant family in the present study is the first-ranked and the most species-rich family in the Flora of Ethiopia and Eritrea (Ermias Aynekulu *et al.*, 2012; Zeleke Assefa *et al.*, 2023). The dominance of the Fabaceae family may be attributed to its taxa having the ability to disperse and adapt to a wide range of ecological and physiological factors that allow them to thrive in different habitats and environmental conditions (Getahun Yemata and Genetu Haregewoien, 2022; Zeleke Assefa *et al.*, 2023).

3.4.2. Plant community types

The finding of four significantly different ($P \leq 0.001$) community types indicates the relative woody plant diversity in the studied forest patches. These plant communities occur in the altitudinal range of 1925 to 2413 m.a.s.l. and are identified with the analytical techniques known as agglomerative hierarchical clustering and ordination method applied to each forest patch. The MRRP results show that there are significant differences ($R = 0.4703$, $P \leq 0.001$) among the four community types. This could be attributed to environmental and anthropogenic factors that cause the variation in species composition among the plant communities (Misganaw Meragiaw *et al.*, 2018; Rahman *et al.*, 2022; Metsehet Yinebeb *et al.*, 2023; Zeleke Assefa *et al.*, 2023).

Although the three forest patches exhibit notable degrees of similarity (Ss Mean ±Sd = 0.65±0.08), it is primarily due to the location and relatively similar operating environmental factors (Friis *et al.*, 2011). The redundancy analysis (RDA) employed in this study revealed significant associations between community types and environmental variables within the Gerado, Micha, and Mekelet forest patches. Altitude, slope and tree cutting were identified as crucial factors influencing the composition and distribution of community types, with varying degrees of significance. Similarly, various researchers in Ethiopia indicated that altitude is a key environmental factor determining the vegetation distribution pattern (Haile Yineger *et al.*, 2011; Misganaw Meragiaw *et al.*, 2018; Admassu Addi *et al.*, 2020; Abera Anamo *et al.*, 2023; Metsehet Yinebeb *et al.*, 2023; Zeleke Assefa *et al.*, 2023).

The results revealed that the species diversity and evenness for each forest patch with the mean value ranging from 1.18 to 2.752 and 0.720 to 0.779 respectively are within the acceptable range of the Shannon-Wiener diversity index (Kent and Coker, 1992). Gerado has the highest species richness (50), followed by Micha (32) and then Mekelet (21). This variation could be attributed to environmental and anthropogenic factors (Abyot Dibaba *et al.*, 2014; Misganaw Meragiaw *et al.*, 2018; Abera Anamo *et al.*, 2023; Zeleke Assefa *et al.*, 2023). The latter is verified by our results from the anthropological analysis that showed the Mekelet forest patch faces the highest disturbance level, primarily driven by the intense cutting and firewood collection observed.

3.4.3. Anthropogenic disturbances

Anthropogenic disturbances including cutting, firewood collection, charcoal production, agricultural expansion, grazing, and browsing are important factors that affect the species distribution and vegetation structure of the forest patches in this study. Many studies indicated that anthropogenic factors often have a pronounced effect on plant species diversity, richness distribution pattern and vegetation structure of forest ecosystems (Yitayih Dagne and Liyew Birhanu, 2023; Zeleke Assefa *et al.*, 2023; Fitsum Temesgen *et al.*, 2021; Fikadu Yirga *et al.*, 2019; Zhang *et al.*, 2017). In Gerado, cutting and agricultural expansion emerge as the primary contributor to disturbance, likely due to factors such as population pressure, agricultural practices, and land-use changes. The moderate impact in Gerado forest patch suggests a balance between human activities and the forest ecosystem, possibly influenced by local agricultural practices. Micha forest patch experiences a comparatively lower overall disturbance, with cutting and agricultural expansion playing important roles. This could be attributed to factors such as lower population density, traditional land-use practices, or conservation efforts in the area, resulting in a more sustainable interaction between the local community and the forest. The Mekelet forest patch faces the highest disturbance, primarily driven by intense cutting and firewood collection. This heightened impact may be influenced by factors such as higher population density, increased demand for resources, or limited alternative livelihood options. Among the anthropogenic factors, charcoal production has varied impacts across forest ecosystems in Ethiopia (Birhanu Bekele and Abdo Kemal, 2022; Ewunetu Tazebew *et al.*, 2023). Charcoal production exhibits diverse impacts across the three forest patches, with Gerado experiencing minimal influence, possibly due to a strong local commitment to protect the forest. In the Micha forest patch, an even lower impact was observed, suggesting conservation efforts, cultural norms, and local initiatives play a role in discouraging or regulating charcoal production. These protective actions by the local community in Gerado and Micha contribute to the preservation of their respective forest patches. In contrast, the Mekelet forest patch shows a moderate impact from charcoal production, potentially driven by factors such as increased population pressure and reliance on charcoal as alternative energy sources.

3.4.4. Vegetation structure

Dodonaea viscosa subsp. *angustifolia, Olea europaea* L. subsp. *cuspidata* and *Vachellia sieberiana* are the most dominant woody species found in the study forest patches. The dominance of *Dodonaea viscosa* subsp. *angustifolia* in Gerado and Micha is a common finding in DAF of Ethiopia and semi-arid African woodlands and may be linked to its adaptability to drier conditions (Harrington and Gadek, 2009; Hadgu Hishe *et al.*, 2021; Mubarek Eshetie *et al.*, 2021). The prevalence of *Vachellia sieberiana* in Mekelet can be compared to its prominence in East African savannas, often associated with overgrazing protection that favors its growth (Mugunga and Mugumo, 2013).

In the forest patches of Gerado, Micha, and Mekelet, the frequency and relative frequency (RF) results indicate a pronounced prevalence of *Dodonaea viscosa* subsp. *angustifolia* and *Vachellia sieberiana*, denoting their ecological dominance within these patches. Mekelet, however, distinguishes itself not only with the absolute frequency of *Vachellia sieberiana* but also with the notable presence of *Eucalyptus camaldulensis*, a non-native species, which suggests differential human impact when compared to the other two patches. This pattern of species frequency aligns with broader ecological observations across Ethiopia, where specific species exhibit dominance due to their resilience to environmental stresses and disturbances, potentially shaping the forest structure (Pohjonen and Pukkala, 1990; Shiferaw Alem and Pavlis, 2012).

The study results revealed that the DBH and height classes of the forest patches had inverted J shape distribution patterns attributed to the stable population status (Birhanu Woldie *et al.*, 2015; Alemayehu Yigeremu and Woldearegay, 2022; Getahun Yemata and Genetu Haregewoien, 2022). The study shows that while younger trees dominate in all sites, there are differences in tree and shrub size distribution. Micha tends to have a slightly higher proportion of very young trees, whereas Mekelet shows a tendency for a more substantial presence in the intermediate-size classes. The slightly higher percentage in Micha could indicate either a more diverse range of young species or a more conducive environment for young tree growth. In this study, the scarcity of trees in the tallest categories (10.1 - 15.0 meters and beyond) across all patches, typically below 2%, suggests a notable absence of mature trees, possibly due to historical harvesting, natural thinning, or other restrictive growth factors. This distribution highlights the potential need for strategies to encourage a more diverse age structure, ensuring ecological balance and sustainable forest conservation. The IVI value indicated that, *Olea europaea* L. *subsp. cuspidata*, *Pittosporum viridiflorum*, *Dodonaea viscosa* subsp. *angustifolia* and *Vachellia sieberiana* are the most important species in the study area. This indicates their dominance and prevalence in terms of density, frequency, and canopy cover within the forest patches.

The population structure pattern of selected woody plant species based on their Importance Value Index (IVI) in the studied forest patches reveals significant ecological insights. *Pittosporum viridiflorum* and *Olea europaea* L. subsp. *cuspidata* exhibit an inverted J-shaped pattern, indicative of healthy regeneration but necessitating further investigation into why fewer individuals reach maturity (Tamrat Bekele. 1993). This could point towards potential issues in the later stages of growth, such as competition or anthropogenic factors. *Dodonaea viscosa* subsp. *angustifolia* shows a broken inverted J-shaped pattern, emphasizing the adverse effects of selective cutting. This highlights the critical need for implementing sustainable resource management practices to ensure long-term viability (Zeleke Assefa *et al.*, 2023). Meanwhile, *Vachellia sieberiana* displays a bell-shaped pattern, suggesting a stable population overall. However, there is a need to focus on the factors that hinder individuals from surviving into the largest diameter at breast height (DBH) classes. Addressing these factors is essential for maintaining the ecological balance and ensuring the persistence of these species within the forest ecosystem (Kflay Gebrehiwot. 2020).

3.5. Conclusion

The present study describes the woody species floristic composition, plant community types, vegetation structure, and anthropogenic disturbances of the Gerado, Micha, and Mekelet forest patches located in northern Ethiopia. The observed low woody species richness in the study area, compared to other dry Afromontane forests in other parts of Ethiopia, underscores the influence of diverse factors such as anthropogenic disturbance, altitude, climate, and environmental heterogeneity on species composition. The dominance of the Fabaceae family aligns with broader observations in the Flora of Ethiopia and Eritrea as this is the family that came up with the highest number of species among the close to 250 or so angiosperm families found in Ethiopia. The identification of four distinct community types within the altitude range of 1925 to 2413 m.a.s.l., coupled with the significant associations between environmental variables and community types, highlights the importance of altitude, aspect, slope, and disturbance in shaping the composition and distribution of plant communities in these forest patches. The three patches were economically and ecologically dominated by *Dodonaea viscosa* subsp. *angustifolia*, *Olea europaea* L. subsp. *cuspidata* and *Vachellia sieberiana*. The prevalence of certain woody species, such as *Dodonaea viscosa* subsp. *angustifolia* and *Vachellia sieberiana*, reflects their adaptability to specific ecological conditions. The study also emphasizes the impact of anthropogenic disturbances, with cutting, firewood collection, and charcoal production influencing species distribution and vegetation structure. The variation in disturbance levels across the three forest patches underscores the complex interplay between human activities, conservation efforts, and ecological sustainability. Overall, these findings contribute to a comprehensive understanding of the ecological dynamics of the studied dry Afromontane forests, offering valuable insights for conservation and management strategies in the face of environmental and anthropogenic challenges.

Recognizing the existing community-based conservation and protection practices within the Gerado, Micha, and Mekelet forest patches, this study highlights the critical need for enhancing and implementing sustainable forest conservation strategies. Such strategies should aim not only to preserve biodiversity but also to effectively mitigate the impacts of anthropogenic and environmental challenges. It calls for a concerted effort among local communities, researchers, and policymakers to build upon current practices, ensuring a comprehensive and informed approach to maintaining the ecological integrity of these vital Dry Afromontane Forest patches.

CHAPTER FOUR

4. Ethnobotanical Study of Traditional Medicinal Plants Used by the Local People in Habru District, North Wollo Zone, Ethiopia

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Ethnobotanical Study of Traditional Medicinal Plants Used by the Local People in Habru District, North Wollo Zone, Ethiopia

Abstract

Background: Ethiopia is a country located in the Horn of Africa, which combines richness in plant resources and cultures of human plant use. The people of Habru District of North Wollo Zone (Amhara Region, Ethiopia) have a long history of use of plant resources for various purposes including in traditional herbal remedy preparation and use. However, the District has not been adequately studied for its ethnobotanical resources and the associated knowledge. This study focused on human medicinal plants and their traditional uses in Habru District. The objective of the study was to document and analyze the plant species used by the local communities to treat human ailments along with the associated traditional knowledge and practices.

Methodology: The study was carried out in Habru District from June, 2021 to December, 2022. Ethnobotanical data were collected using semi-structured interviews, guided field walks, 13 focus group discussions (one at the district level and 12 at the kebele/subdistrict level) and market surveys. A total of 388 informants (250 males and 138 females) were selected from all 13 kebeles within Habru District using systematic random sampling, and 42 key informants were purposively selected. Descriptive statistics, preference ranking, direct matrix ranking, informant consensus factor and fidelity level were applied for data analysis.

Results: The results provide insights into the medicinal plant diversity within Habru District, where 134 plant species in 110 genera and 54 families were documented, including 2 endemics, highlighting the district's significance in biodiversity conservation and healthcare delivery. Disease prevalence analysis showed that gastrointestinal and parasitic ailments (ICF = 0.85), febrile diseases (ICF = 0.84), and culture-related conditions exhibit high informant consensus factors. Remedy preparation involves various plant parts, predominantly leaves (47.3%), followed by roots (22.1%), fruits (7.0%), and seeds (5.8%). Freshly harvested plant parts were frequently used (58.2%), while 24.7% involved both dried and fresh parts. Oral application (47.3%) and topical use (31.8%) are the major routes of remedy administration. The marketability of medicinal plants was evident, with 16.4% of the species reported as marketable, including *Terminalia brownii* Fresen. *Myrtus communis* L., *Ruta chalepensis* L., *Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif., *Allium sativum* L. and *Capsicum annuum* L.. Multipurpose plants such as *Solanum somalense* Franchet. (91.3% FL), *Ocimum lamiifolium* Hochst. ex. Benth. (88.9% FL), and *Verbascum sinaiticum* Benth. (85.7% FL) exhibited notable healing potentials.

Conclusion: The current study underscores the intricate relationship between the local community and medicinal plants, emphasizing the importance of biodiversity conservation and healthcare, and acknowledging the dynamic interplay between cultural heritage and ecosystem health. The results contribute to development of sustainable conservation strategies, healthcare practices and the preservation of traditional knowledge, and highlight the

interdependence of human societies and their natural environments. Community-based conservation initiatives with active participation of local communities are desirable for the conservation and sustainable use of medicinal plant species and their habitats. Raising public awareness about the sustainable harvesting and utilization of marketed medicinal plants (e.g., *Terminalia brownii* Fresen.) that are under threat is also important to ensure their availability for future generations and contribution to socioeconomic development.

Keywords: Biodiversity conservation, Ethnobotany, Habru District, Traditional herbal knowledge, Traditional medicinal plants

4.1. Background

Medicinal plants are vital resources for primary healthcare of people around the world (Ekor, 2014). Approximately 80% of the global population is estimated to utilize medicinal plants for disease treatment, and in African nations, this percentage is even higher (WHO, 2001; Mahomoodally, 2013; Gakuya *et al.*, 2020). Ethiopia is a renowned hub for ethnomedicinal research due to its remarkable plant diversity, cultural richness and profound traditional knowledge and ancient medical practices (Tilahun Teklehaymanot and Mirutse Giday, 2010; Mersha Ashagre and Ermias Lulekal, 2021). Among the twelve Vavilov centers of origin, Ethiopia exhibits an enormous diversity of domesticated crops and their wild relatives, demonstrating a vast array of plant genetic resources (Vavilov, 1951; Westphal, 1974). The flora of Ethiopia is estimated to contain close to 5,757 vascular plant species, with approximately 10% endemic to the flora area (Sebsebe Demissew *et al.*, 2021; Ensermu Kelbessa and Sebsebe Demissew, 2014). In the healthcare system of Ethiopia, traditional medicine is widely practiced alongside modern medical approaches (Lemessa Etana *et al.*, 2019; EPHI, 2023). About 80% of Ethiopian population rely on traditional medicines (TMs) for their primery healthcare, and more than 95% of the preparations are made from plants (Nigatu Tuasha *et al.*, 2018; Lemessa Etana *et al.*, 2019). However, these medicinal plants face various threatening factors, including habitat destruction, urbanization, agricultural expansion, deforestation, firewood collection, and environmental degradation (Mirutse Giday *et al.*, 2010; Ermias Lulekal *et al.*, 2013; Admasu Moges and Yohannes Moges, 2020; Moa Megersa and Samuel Woldetsadik, 2022).

Various ethnobotanical studies have been conducted in different parts of Ethiopia to document the extensive use of medicinal plants to treat human and livestock ailments (Ermias Lulekal *et al.*, 2013; Getnet Chekole, 2017; Tilahun Teklehaymanot, 2017; Gadisa Demie *et al.*, 2018; Leul Kidane *et al.*, 2018; Banchiamlak Nigussie and Kim, 2019; Tibebu Tefera and Mesele Yihune, 2019; Netsanet Gonfa *et al.*, 2020; Zewdie Kassa *et al.*, 2020; Muhidin Tahir *et al.*, 2021; Yimer Assen *et al.*, 2021; Melaku Masresha *et al.*, 2022; Melese Bekele *et al.*, 2022; Moa Megersa and Samuel Woldetsadik, 2022). Despite the crucial role of medicinal plants in Ethiopia's traditional primary healthcare, the geographical and cultural coverage is limited and so are attempts to scientifically explore, document and validate the depth of the associated knowledge (Moa Megersa and Samuel Woldetsadik, 2022).

Similar to elsewhere in Ethiopia, people living in Habru District have also traditional practices to take care of themselves and the health of their livestock (HDARDO, 2019). Additionally, the ecology of Habru District is characterized by highlands, middle lands, and lowlands. Due to the diverse ecological landscape, the diversity and practices of traditional MP species are expected to be more in the study area. Therefore, a comprehensive ethnobotanical study in Habru district is essential to document and analyze the traditional knowledge and practices of the local people focusing on medicinal plants for the treatment of human ailments, contributing to the utilization and preservation of biodiversity. Moreover, comparing the findings of this study with the Ethiopian ethnobotanical medicinal plant database will provide valuable insights into the regional distribution and utilization of medicinal plants, further enriching our understanding of Ethiopia's remarkable traditional plant-based healthcare system. In view of this, the present study aims to (i) collect, identify, and document medicinal plants and the associated indigenous and local knowledge of the people used to treat various human ailments in the study area (ii) identify and document marketable medicinal plants used in the study area and (iii) select candidate MP species of high informant consensus and fidelity level values for antimicrobial and phytochemical analyses in our subsequent studies.

4.2. Materials and methods

4.2.1. Description of the study area

The study focuses on the ethnobotany of medicinal plants in the Habru District, situated in the North Wollo Zone of the Amhara Region, Ethiopia (Figure 3.1). Habru District has 36 rural kebeles (subdistricts) and three urban administrations, with a total area of 1350.4322 km^2. According to the Habru District Agriculture and Rural Development Office 2019 report, Mersa town is the center of the district, which is 88 km north of Dessie town and 491 km north of Addis Ababa, the Ethiopian capital, 406 km east of Bahirdar (Amhara Region’s capital), 30 km northeast of Woldiya (North Wollo Zone capital) (HDARDO, 2019). The grid references for Harbu District come within 11°35'0"-11°55'0"N and 39°30'10"- 40°10'0"E with an altitudinal range of 1430-2800 masl. It is bordered to the south by the Mille River, separating it from the South Wollo Zone. To the west lies Gubalafto District, while the Alawuha River forms the northern boundary, separating North Wollo from Kobo woreda. The eastern border is defined by the Afar Region. The weather condition of Habru District is characterized by the cold climate which is locally known as DEGA (temperate) and covers 3.5% of the total area of the district, the warmest, which is locally known as KOLLA (tropical) and covers 56.5% and medium, which is locally known as WEINADEGA (subtropical) and covers 40% (HDARDO, 2019). Rainfall data from 1986-2019 was obtained from the Ethiopian National Meteorology Agency (ENMA) Sirinka station, the mean annual temperature of the study area is 20.1 °C (Figure 3.2). This corresponds to the monthly minimum and maximum temperatures of 10.6 °C and 30.8 °C respectively (ENMA, 2019). Habru District receives a bimodal rainfall of about 1045 mm/year in which high and low rainfall was received during 'Kiremt' (June to October) and 'Belg' (March and April) respectively. The district has also a population of 192,742, with a majority residing in rural areas (HDARDO, 2019). The population density is 155.46 persons per square kilometer, higher than the zone average. The inhabitants mainly practice Muslim (76.85%) and Ethiopian Orthodox Christianity (22.95%) (HDARDO, 2019). The economic activities in Habru District are centered around large commercial farms focused on livestock rearing and agro-processing, particularly in the Girana kebele. The district benefits from market access facilitated by the main road connecting it to Addis Ababa, and local products are sold in nearby markets such as Mersa and Dessie town.

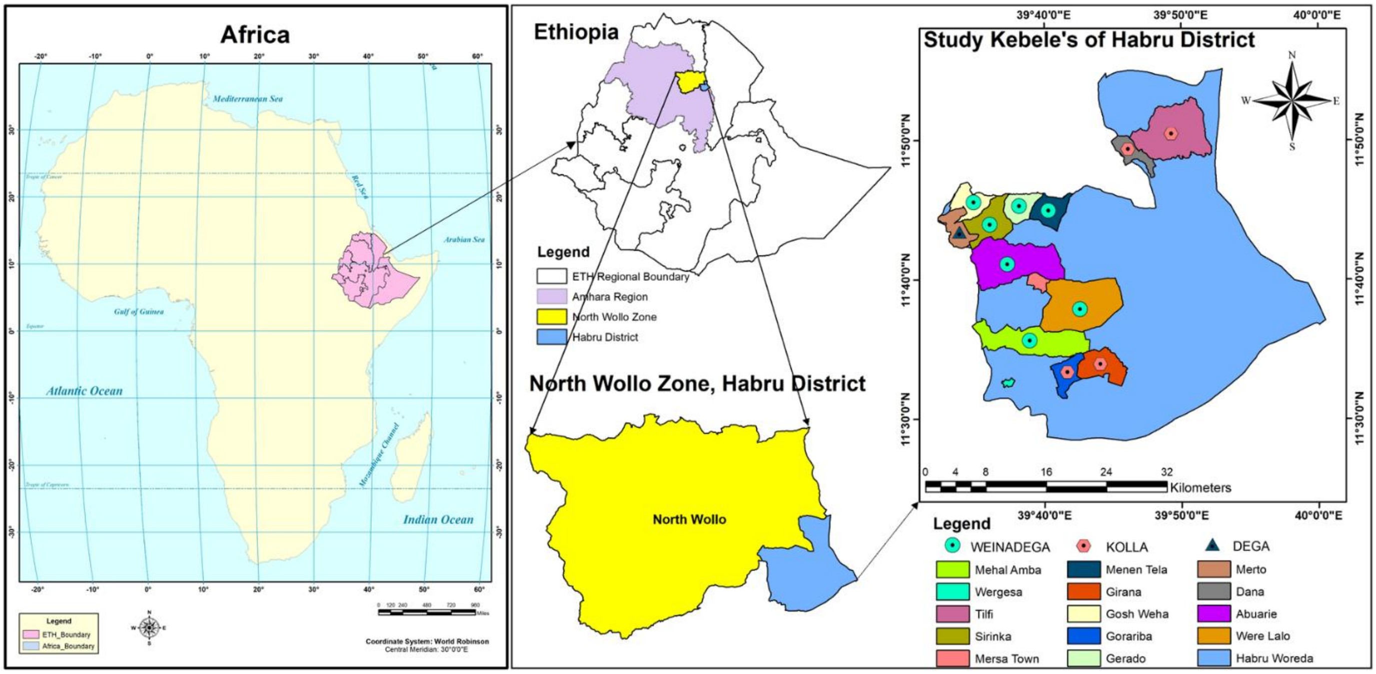


Figure 3.1: Map of Ethiopia showing the Amhara region and the location of the study district (developed using ArcGIS 10.5)

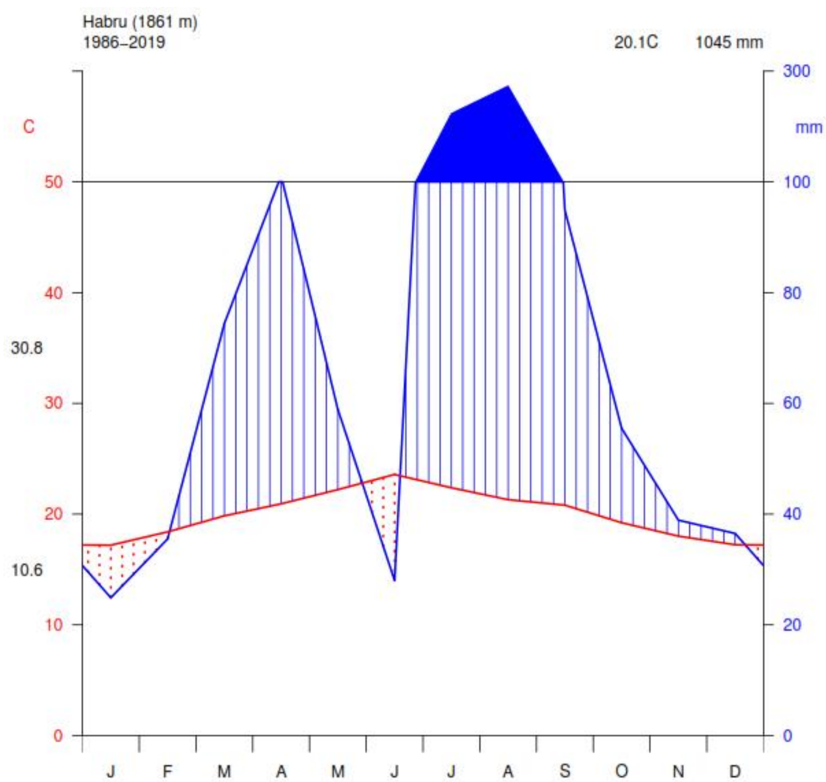


Figure 3.2: Climate diagram of the Habru District showing rainfall distribution and temperature variation from 1986 - 2019. Data source: Ethiopian National Meteorology Agency (ENMA**)**

4.2.2. Reconnaissance survey and site selection

Before the commencement of the reconnaissance survey, an official letter was received from the Department of Plant Biology and Biodiversity Management (DPBBM) at Addis Ababa University. Additionally, verbal informed consent was obtained from each participant involved in the study during the designated period. The reconnaissance survey took place between June 14, 2021, and July 02, 2021, within the confines of Habru District. Its purpose was to develop an initial understanding of the agro-ecological characteristics of the region, the prevailing vegetation status, the local population's indigenous and local knowledge regarding plant applications for various uses, and an assessment of accessibility and other pertinent environmental conditions. To select study kebeles, the purposive sampling method was employed, with a focus on identifying kebeles with better vegetation cover and a well-known history of utilizing medicinal plants. These kebeles were also deemed potential sites for home gardening activities. In addition, the selection of study sites was underpinned by prior insights gathered from participants in focus group discussions (FGDs), community leaders, venerable elders, traditional healers, and healthcare personnel within the locality. As a result, a total of 13 study kebeles were chosen, representing 35% of the entire kebele count within Habru District. These selected study kebeles are enumerated as follows: Worgesa, Gosh Weha, Sirinka, Gerado, Girana, Dana, Abuarie, Tilfi, Menen Tela, Merto, Gorariba, Mehal Amba, and Were Lalo.

4.2.3. Sample size determination and selection of informants

The determination of the informant sample size adhered to the methodology outlined in Cochran (1977) as cited in Bartlett et al. (2001). Accordingly, the sample size (n) was calculated using the formula:

$$\mathrm{n} = \frac{N}{1 + \mathrm{N}\ (\mathrm{e})^2}$$

where **n** =sample size to be used for this research, **N** = total number of households (HH) in all sub-districts, **e** = maximum variability or margin of error 5% (0.05), **1** = the probability of the event occurring. To establish the number of households to be interviewed within each kebele, the following formula was applied:

$$A = \frac{B\,x\,n}{N}$$

where **A** = **s**ample size to be used in a given kebele, **B** = total number of households (HH) found in a given kebele, **n** = total sample size to be used for this research, and **N** = total number of households (HH) in all sub-districts.

The research encompassed a total of 388 informants (250 males and 138 females) from all 13 kebeles within Habru District (Table 3.1). The selection of these informants followed systematic random and purposive sampling methods, including peer recommendations, as described by (Martin, 1995). Nominations of traditional herbalists to participate as key respondents were collected from community members, elderly people and knowledgeable inhabitants as the methods recommended by Alexiades (1996); Cotton (1996) and used to identify 42 key informants (34 men and 8 women) among the inhabitants, whereas general informants were systematically sampled during random visits made to houses in the study kebeles.

Table 3.1: Description of kebeles (subdistricts) of data collection within the study area, highlighting key geographical and demographical attributes.

Study kebele/village name	GPS coordinates		Elevation (m)	Ecology	Total number of HH	Number of interviewees	Gender		Age		Occupation		Religion	
	Latitude	Longitude					M	F	YM	El	Li	Il	Ch	Mu
Wergesa	11°32'37.38"N	39°37'20.53"E	1848	SuT	1600	48	24	24	18	30	19	29	23	25
Gosh Weha	11°45'17.86"N	39°34'45.29"E	2316	SuT	663	20	13	7	5	15	7	13	12	8
Sirinka	11°44'1.77"N	39°35'59.59"E	2111	SuT	780	24	18	6	7	17	5	19	21	3
Gerado	11°45'5.36"N	39°38'2.04"E	1817	SuT	674	20	8	12	6	14	5	15	11	9
Girana	11°34'0.43"N	39°44'8.96"E	1425	Tro	1301	39	22	17	15	24	8	31	4	35
Dana	11°49'28.33"N	39°46'17.45"E	1545	Tro	734	22	16	6	6	16	11	11	0	22
Abuarie	11°41'14.17"N	39°36'49.34"E	1811	SuT	1707	51	36	15	9	42	17	34	41	10
Tilfi	11°50'31.75"N	39°49'21.32"E	1534	Tro	729	22	19	3	11	11	3	19	0	22
Menen Tela	11°44'52.20"N	39°40'29.71"E	1749	SuT	585	18	11	7	1	17	3	15	10	8
Merto	11°43'26.68"N	39°33'41.16"E	2589	Tem	1000	30	20	10	11	19	7	23	22	8
Gorariba	11°33'7.89"N	39°41'39.22"E	1547	Tro	514	16	13	3	2	14	6	10	2	14
Mehal Amba	11°35'36.91"N	39°39'5.25"E	1645	SuT	1966	59	35	24	22	37	23	36	9	50
Were Lalo	11°38'11.28"N	39°42'41.59"E	1449	SuT	632	19	15	4	3	16	5	14	2	17
				Total	**12885**	**388**	**250**	**138**	**116**	**272**	**119**	**269**	**157**	**231**

Key: **Household**, HH; **Ecology** (Subtropical, SuT; Tropical, Tro and Temperate, Tem); **Gender** (Male, M and Female, F); **Occupation** (Literate, Li denotes interviewees who have completed at least primary education and Illitrate, Il); **Age** categories include young to middle-aged, YM (19–39 years) and elderly, El (40–98 years). **Religion** (Christian, Ch encompasses orthodox, catholic, and protestant and Muslim, Mu). Regarding ethnicity and language, all interviewees belong to the Amhara ethnic group and speak the Amharic language.

4.2.4. Data collection

The ethnobotanical data collection encompassed three distinct field trips carried out between June 14, 2021, and December 14, 2022. Qualitative and quantitative ethnobotanical data were collected from informants through a pre-prepared semi-structured interview method (Martin, 1995; Alexiades, 1996; Cotton, 1996). Notably, field notes were meticulously recorded, safeguarding the discreet knowledge of the local community (Martin, 1995). To ensure confidentiality, necessary ethical clearance was secured by briefing the informants, and the fieldwork adhered to Bennett's Golden Rules (McClatchey and Gollin, 2005). In the broader context of this study, techniques such as group discussions, semi-structured interviews, guided field walks, market surveys, preference ranking, and direct matrix ranking were employed, as recommended by (Martin, 1995).

Focus group discussion and semi-structured interview

Semi-structured questionnaires were employed to facilitate discussions and interviews with informants, enabling the collection of pertinent information aligned with the research objectives. The methods, procedures, and techniques followed those recommended by standard ethnobotany sources (Alexiades, 1996). Additionally, a total of 13 focus group discussions were carried out, one at the district level and 12 at selected kebeles of the district. In each focus group discussion key informants, traditional healers, elders, kebele and district administrative officials from natural resource and forest protection offices, and agricultural and rural development offices were involved to amplify insights into medicinal plant knowledge at the community level and to corroborate information obtained through semi-structured interviews (Martin, 1995). Informants were interviewed separately in the local Amharic language, addressing queries about their general information. Furthermore, interviewees provided local names of medicinal plants, detailed information about treated ailments, species habitats, seasonality, marketability, plant parts used, condition of those parts (fresh or dried), preparation methods, dosage instructions, and routes of administration for remedies (Martin, 1995; Cotton, 1996). Moreover, informants were asked about other (non-medicinal) uses of the medicinal plant species they mentioned to identify the overall use values and use diversities of species in the manner recommended by Martin (1995); Alexiades (1996) and Cotton (1996).

Guided field walk

Field observations were facilitated by the collaboration of local guides, traditional healers, district experts, and participating informants, ensuring the comprehensive acquisition of indispensable data within the study area. Through the integration of both etic and emic categorizations, invaluable insights were garnered, shedding light on the categorization of medicinal plants, plant communities, landscapes, and soil types. Additionally, the pivotal contribution of traditional healers during the guided field walk extended beyond facilitating the process; they actively participated in identifying encountered medicinal plants by providing their vernacular names, medicinal use, parts used, preparation methods, dosages, and methods of application. The collection of voucher specimens during the guided field walk was supported by digital photographs of both freshly gathered and pressed dry voucher specimens. This specimen collection endeavor encompassed diverse environments, including the wild, home gardens, crop fields and margins and local markets.

Market survey

Market surveys were carried out to document the medicinal plants available in local markets of the study area and gather insights into the market values of plants. This method proved instrumental in investigating the availability, pricing, and unit measurement of marketable medicinal plants, aiding the preservation of high-value medicinal plant species. To this end, four local markets namely Mersa, Mehal Amba, Wergesa and Girana were visited, and semi-structured interviews were conducted with drug vendors at the markets. A total of 18 informants (12 vendors and 6 user of MPs) were interviewed, with 7 men and 11 women, representing participants from all four local market areas of the study. These interviews aimed to obtain general information on the multipurpose roles and marketability of medicinal plant species, and their plant-derived products sold on markets were recorded.

Voucher specimen collection and identification

Voucher specimens of reported medicinal plants were collected and the preliminary identification was executed using manuals in the field and they were pressed, dried, deep frozen, and identified. The identification was performed using the keys from published volumes of the Flora of Ethiopia and Eritrea, followed by comparisons with authenticated specimens in the National Herbarium (ETH) of Addis Ababa University, and ultimately confirmed by taxonomic experts at Addis Ababa University. The identified specimens with voucher numbers, families, species, and vernacular names, dates and sites of collection were recorded and deposited at the National Herbarium (ETH) at AAU.

4.2.5. Data analysis

For this study, a combination of qualitative and quantitative ethnobotanical analytical tools was employed, adhering to the relevant methodologies proposed by Martin (1995) and Cotton (1996). Ethnobotanical data were entered into an Excel spreadsheet, version 2013, and subjected to comprehensive analysis using descriptive statistics. This facilitated the identification of the most frequently employed multipurpose plants within the study area. To elucidate the proportions of different plant species, growth forms, sources of collection, plant parts used, methods of preparation, and related aspects, a range of ethnobotanical scoring and ranking techniques, percentage frequency methodologies, and inferential statistics were employed. The findings were subsequently presented through graphs, charts, tables, and textual explanations.

The preference ranking (PR) technique involved the systematic arrangement of lists or groups of plants or resources based on a designated criterion (Martin, 1995). In this study, key informants were requested to assign values or scores for ranking the most preferred plant for treating the most reported human ailments within the study area. Each informant ranked the items according to individual preference or perceived importance within the community. The items were assigned numerical values, with the most vital receiving the highest number, descending in value as the significance of the items diminished. The least preferred or important item was denoted by the lowest value, which was '1'.

Direct matrix ranking (DMR) was conducted in line with the method described by (Martin, 1995), aiming to compare the multipurpose use of medicinal plants. The process involved soliciting informants to sequentially order a given set of items according to specific attributes. This procedure was applied to ten multipurpose medicinal plants and the five most cited factors perceived as threatening by key informants, following Cotton (1996). Each informant provided use values ranging from 5 for "excellent" to 0 for "not used".

The informant consensus value was calculated based on the free listing data collected during interviews, which were then summarized in tabular form (Phillips, 1996). The level of agreement between information provided by various informants was assessed using the Informants' Consensus Factor (ICF) formula (Trotter and Logan, 1986):

$$ICF = \frac{Nur - Nt}{(Nur - 1)}$$

where $N_{ur,}$ is the number of use reports from informants for a particular plant usage category, and N_t is the number of species that are used for the plant usage category for all informants. Values range between 0 and 1, where 1 indicates the highest level of informant consent. As described by (Heinrich *et al.*, 1998), medicinal tradition is viewed as well-defined if a high degree of consensus is recorded. This means that a high value indicates that relatively few taxa are used by a larger proportion of the healers, while a low value indicates that informants disagree on the taxa to be used in the treatment within a category of illness.

Fidelity Level (FL) has been employed to quantify the importance of a given species for a particular purpose in each cultural group (Friedman *et al.*, 1993) cited in Alexiades (1996). It is the percentage of informants claiming the use of a certain plant species for the same major purpose, and FL was calculated for the most frequently reported diseases or ailments as follows:

$$\text{FL (\%)} = \left(\frac{Np}{N}\right) x\ 100$$

where Np is the number of informants that claim the use of plant species to treat a particular disease; and N is the number of informants that use the plants as a medicine to treat any given disease.

Knowledge difference according to demographic characteristics of informants

For this study, ethnobotanical knowledge exhibits distinct variations based on the diverse demographic characteristics of the informants. Male general informants, constituting 62.4% of the participants, reported about the use of plants as medicine in the study area, while the key informants were predominantly masculine (81%). The study cohort encompassed a wide age range, with 29.9% falling into the young-to-middle-aged category (19–39 years old) and 70.1% comprising the elderly individuals (40–98 years). Notably, 30.7% of the informants displayed literacy, while the majority remained illiterate (69.3%). Regarding marital status, the majority of informants were married (85.3%), followed by divorced (7.2%), widowed (4.4%), and single

(3.1%) household representatives. This intricate demographic mosaic contributes to the research's holistic scope, ensuring comprehensive representation across gender, age, literacy levels, and marital statuses.

4.3. Results

4.3.1. Diversity of reported medicinal plants in Habru District

A total of 134 medicinal plant species belonging to 110 genera and 54 botanical families consisting of 132 angiosperms and 2 gymnosperms were reported to be used for treating human ailments in Habru District (Figure 3.3 and Table 3.2). Of these plants, herbs took the highest proportion (36%) whereas climbers took the least proportion (Figure 4). The family Solanaceae was represented by the highest number of species (12 species, 9%) followed by Fabaceae (10 species, 7.5%) and Lamiaceae (7 species, 5.2%). Asteraceae, Cucurbitaceae, Euphorbiaceae, and Malvaceae each contributed 5 species (3.7% of the total). In contrast, Anacardiaceae, Myrtaceae, and Verbenaceae each represented with 4 species. The top 15 medicinal plant species identified in the study area based on the use citation, compared to similar studies conducted in another region of Ethiopia are presented in Table 3.3.

Among the identified MPs in Habru District, indigenous species hold the predominant position, constituting 107 (79.9%) of the total. These plants demonstrate adaptation to the local ecosystem over time, reflecting their deep-rooted connection to the district. Additionally, the district's plant diversity is enriched by the presence of 23 introduced species (17.2%), offering the potential for therapeutic options. Among the reported MPs, two (1.5%) endemic plant species, *Millettia ferruginea* (Hochst.) Hochst. ex Baker and *Urtica simensis* Hochst. ex A.Rich. were recorded in the study area. These species have IUCN conservation statuses of Least Concern (LC) and Not Assessed (NA), respectively.

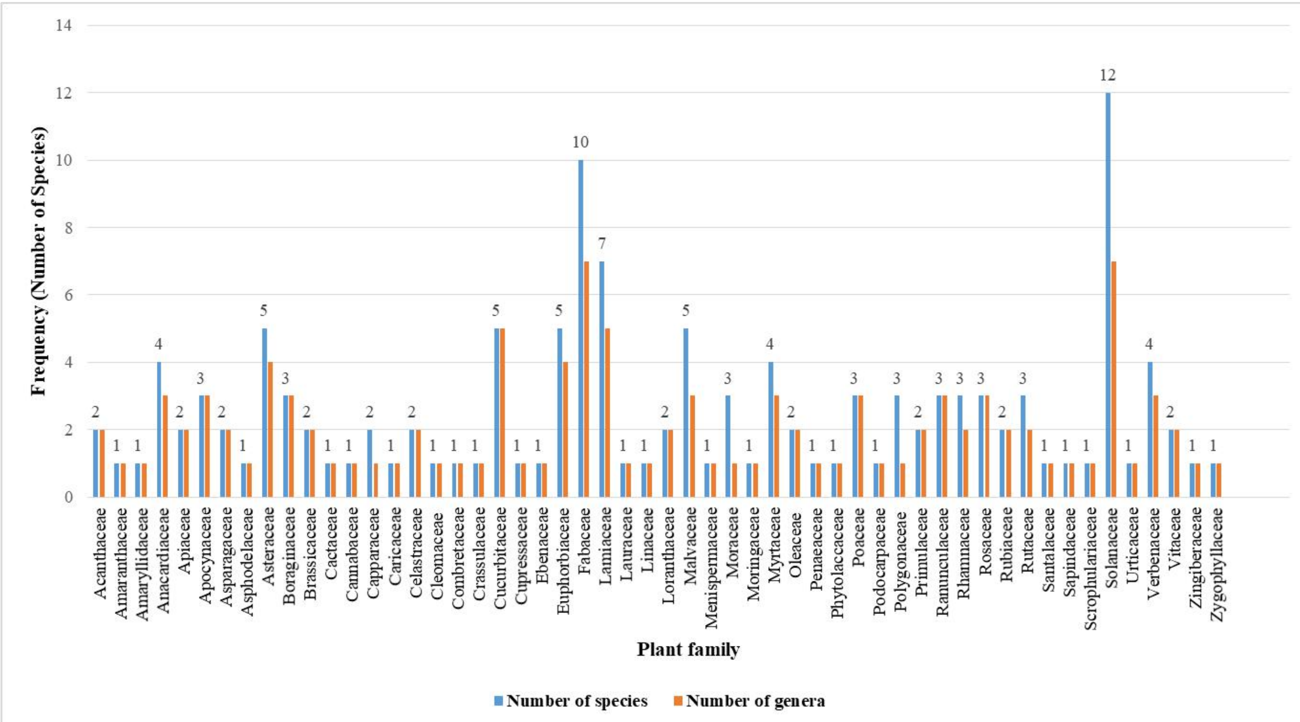


Figure 3.3: Distribution of reported medicinal plant species in Habru District across different families

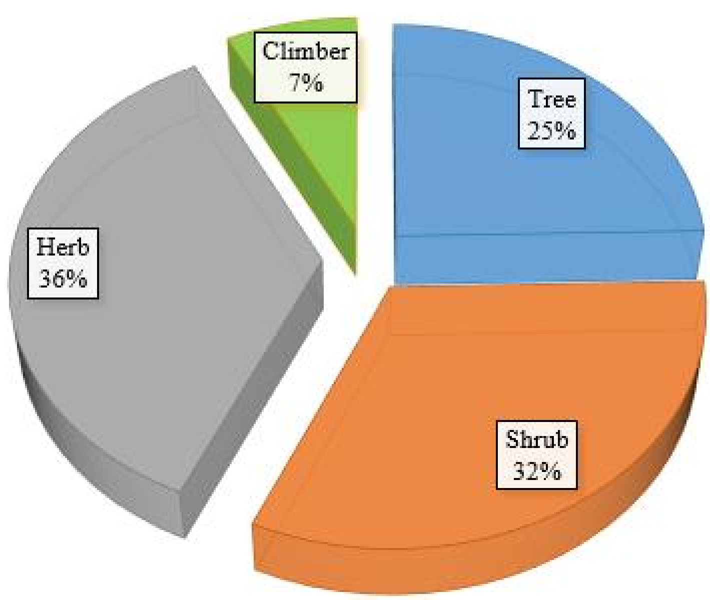


Figure 3.4: Growth form of medicinal plants collected in Habru District

Table 3.2: List of medicinal plants used for the treatment of human ailments in Habru District, Amhara Region, Ethiopia: scientific name; family; local name; growth form; ailment treated; plant parts used; condition of plant part uses; methods of preparation and application, route of administration, plant part mixed with and voucher number

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Barleria eranthemoides R.Br.ex C.B.Clarke	Yeset Af	Acanthaceae	H	Boils	L	F	3	De	2	MA13
				Diarrhea	R	F	1	O	1	
				Wound	R	F	2	De	3	
Justicia schimperiana (Hochst. ex Nees) T.Anderson	Sensel, Simiza	Acanthaceae	S	Febrile disease	L	F	10	ODN	6	MA62
				Typhoid	L	F	16	O	5	
				Malaria	L	F	16	O	2	
				Liver problem	L	F	5	O	2	
Agave sisalana Perrine	Chiret, Kacha	Asparagaceae	S	Wound	Lat	F	4	De	2	MA06
Allium sativum L.	Nech Shnkurt	Amaryllidaceae	H	Atopic eczema	Bu	F	4	De	3	MA08
				Asthma	Bu	F	4	De	3	
				Common cold	Bu	DF	11	O	6	
				Dandruff	Bu	DF	1	O	2	
				Pneumonia	Bu	F	4	De	1	
				Coughing	Bu	DF	1	O	5	
				Malaria	Bu	DF	11	O	1	
Aloe macrocarpa Tod.	Iret / Iret tafa	Asphodelaceae	H	Gastritis	L	F	11	O	2	MA09
				Stomachache	L	F	11	O	2	
				Malaria	Lat	F	11	O	1	
				Wound	Lat	F	4	De	2	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Achyranthes aspera L.	Telenj	Amaranthaceae	H	Nasal infection	R	F	7	Na	4	MA05
				Minor bleeding	L	F	5	O	2	
				Wound	L	F	5	O	2	
Searsia retinorrhoea (Steud. ex Oliv.) Moffett	Talo/Talo Embis	Anacardiaceae	T	Stomachache	R	D	5	O	6	MA96
				Tonsillitis	L	F	5,11	O	10	
Schinus molle L.	Kundo-berberie	Anacardiaceae	T	Common cold	L	F	8	Na	3	MA107
				Jaundices	Fr	D	5	O	2	
Searsia pyroides (Burch.) Moffett	Yeahya Talo, Yeregna Qolo	Anacardiaceae	S	Wound	L	D	3	De	1	MA97
Mangifera indica L.	Mango	Anacardiaceae	T	Impotency	Fr	F	9	O	2	MA72
Ferula communis L.	Dog	Apiaceae	H	Impotency	R	D	13	O	2	MA49
Foeniculum vulgare Mill	Ensilal	Apiaceae	H	Headache	R	F	5	O	3	MA53
				Stomachache	R	D	5	O	2	
Carissa spinarum L.	Agam	Apocynaceae	S	Diarrhea	R	DF	5	O	2	MA22
				Constipation	R	DF	5	O	1	
				Evil spirit	R	DF	8	Na	2	
				Headache	L	F	4	De	1	
				Snake bite	L	F	9	O	3	
Calotropis procera (Aiton) W.T.Aiton	Tobia	Apocynaceae	S	Atopic eczema	Lat	F	4	De	2	MA15
				Hemorrhoid	Lat	F	4	De	3	
				Hemorrhoid	R	F	4	An	4	
Periploca linearifolia Quart.-	Moider	Apocynaceae	C	Diabetes	LR	F	1	O	1	MA111

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Dill. & A. Rich.										
Asparagus africanus Lam.	Yeset Kest, Kestanicha	Asparagaceae	H	Skin diseases	R	F	12	De	1	MA133
Bidens pilosa L.	Yeseytan Merfe	Asteraceae	H	Snake bite	L	DF	5	O	2	MA14
				Wound	L	F	3	De	3	
				Evil spirit	Wp	DF	7	ODN	4	
Artemisia absinthium L.	Arity, Natra	Asteraceae	H	Evil spirit	L	F	8	Na	2	MA10
Guizotia abyssinica (L.f.) Cass.	Nug	Asteraceae	H	Gastritis	Se	D	9	O	2	MA57
Gymnanthemum amygdalinum (Delile) Sch.Bip.	Girawa	Asteraceae	T	Stomachache	L	F	5	O	2	MA122
				Giardia	L	DF	15	O	2	
				Wound	L	D	2	De	1	
Artemisia abyssinica Sch.Bip. ex Oliv. & Hiern	Chikugn	Asteraceae	H	Evil Eyes	Wp	F	8	Na	3	MA11
Balanites aegyptiaca (L.) Delile	Bedena	Zygophyllaceae	T	Stomachache	L	F	5	O	1	MA12
				Dandruff	L	F	4	De	2	
Ehretia cymosa Thonn.	Wulaga	Boraginaceae	T	Evil spirit	Hp	F	7	Na	28	MA42
Cordia africana Lam.	Wanza	Boraginaceae	T	Atopic eczema	L	F	4	De	1	MA34
Cynoglossum coeruleum Hochst. ex A.DC.	Fikru-tena, Chigogot	Boraginaceae	H	Febrile disease	L	F	7	NA	1	MA132
Lepidium sativum L.	Feto	Brassicaceae	H	Tonsillitis	Se	DF	5	O	3	MA67
				Gingivitis	Se	DF	6	O	2	
				Fever	Se	DF	1	O	1	
				Wound	Se	DF	3	De	2	
				Boils due to bacteria	Se	DF	3	De	2	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				S. aures						
Sisymbrium officinale (L.) Scop.	Yewef Gomen	Brassicaceae	H	Skin diseases	L	F	3	De	2	MA109
Opuntia ficus-indica (L.) Mill.	Beles, Qulqual	Cactaceae	S	Coughing	Hp	F	5	O	2	MA86
Capparis tomentosa Lam.	Gimero	Capparaceae	S	Evil spirit	R	DF	7	Na	3	MA18
				Evil Eyes	R	DF	7	Na	2	
Cleome gynandra L.	Abethoye	Cleomaceae	H	Wound	L	F	4	De	5	MA31
				Ear infection	L	F	2	Au	10	
				Evil spirit	L	DF	11	O	12	
Capparis decidua (Forssk.) Edgew.	Kontir	Capparaceae	S	Anthrax	Se	D	5	O	2	MA17
Carica papaya L.	Papaya	Caricaceae	T	Gastritis	Fr	F	9	O	3	MA21
Catha edulis (Vahl) Endl.	Chat	Celastraceae	H	Intestinal parasite	L	F	11	O	1	MA23
Gymnosporia senegalensis (Lam.) Loes.	Atat	Celastraceae	S	Eye disease	Fl	F	3	Op	1	MA73
				Evil spirit	R	D	5	O	2	
				Sexual impotency	SB	D	9	O	3	
Terminalia brownii Fresen.	Wyba	Combretaceae	T	Wound	SB	D	7	Na	1	MA116
Kalanchoe petitiana A.Rich.	Endahula	Crassulaceae	H	Tonsillitis	R	F	8	Na	4	MA63
				Stomachache	R	F	9	O	6	
				Swelling	L	F	12	De	3	
Cucumis ficifolius A.Rich.	Yemidir Embway	Cucurbitaceae	H	Evil spirit	L	DF	7	Na	1	MA36
				Rabies	R	D	1	O	2	
				Stomachache	R	F	5	O	2	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				Wound	L	F	3	De	4	
				Febrile disease	L	F	10	ODN	3	
Zehneria scabra (Linn.f.) Sond.	Hareg resa/Etse Sabieq	Cucurbitaceae	H	Febrile disease	L	F	10	ODN	3	MA125
				Wound	L	F	4	De	2	
				Liver problem	L	F	5	O	1	
Cucurbita pepo L.	Duba	Cucurbitaceae	C	Tap worm	Se	D	9	O	1	MA37
				Ear lesion	Fl	F	2	Au	2	
Lagenaria siceraria (Molina) Standley	Qil	Cucurbitaceae	C	Liver problem	L	F	5	O	1	MA64
Momordica foetida Schumach.	Yekura Hareg	Cucurbitaceae	C	Wound	L	F	3	De	2	MA75
				Coughing	Wp	DF	7	Na	3	
Juniperus procera Hochst. ex Endl.	Yehabesha Tsid	Cupressaceae	T	Fire burn	SB	D	2	De	2	MA61
Euclea racemosa L.	Dedeho	Ebenaceae	S	Wound	L	F	3	De	19	MA46
Croton macrostachyus Hochst. ex Delile	Bisana	Euphorbiaceae	T	Atopic eczema	L	F	4	De	4	MA35
				Liver problem	B	DF	5	O	2	
				Stomachache	B	D	15	O	2	
				Gonorrhea	B	F	5	O	3	
				Malaria	Fr	F	13	O	1	
				Chronic skin diseases	L	F	4	De	4	
				Scabies	L	F	4	De	4	
				Wound	L	F	4	De	3	
				Minor bleeding	L	F	4	De	2	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				Febrile disease	L	DF	7	Na	1	
Euphorbia tirucalli L.	Kinchibt	Euphorbiaceae	S	Wound	Lat	F	4	De	3	MA48
Ricinus communis L.	Gulo	Euphorbiaceae	S	Stomachache	R	DF	7	ODN	2	MA98
				Ear mites	L	F	3	Au	3	
				Hemorrhoid	L	F	4	De	1	
				Scabies	Fr	F	4	O	2	
				Evil spirit	R	F	5	O	1	
Euphorbia abyssinica J.F.Gmel.	Kulkual	Euphorbiaceae	S	Hemorrhoid	Lat	F	4	De	1	MA47
				Take out spine	Lat	F	4	De	2	
Tragia brevipes Pax	Aleblabit, Ablalit	Euphorbiaceae	H	Stomachache	L	F	5	O	1	MA134
Vachellia seyal (Delile) P.J.H.Hurter	Wacho Grar	Fabaceae	T	Wound	L	F	3	De	2	MA03
Trigonella foenum-graecum L.	Abish	Fabaceae	H	Gastritis	Se	D	5	O	1	MA118
Pterolobium stellatum (Forssk.) Brenan	Kentafa	Fabaceae	S	Asthma	B	D	2	Na	2	MA94
Cicer arietinum L.	Shimbira	Fabaceae	H	Malaria	Se	D	11	O	1	MA26
Millettia ferruginea (Hochst.) Bak.	Birbira	Fabaceae	T	Skin infection	Fr	D	4	De	1	MA74
Senegalia senegal (L.) Britton	Sibansa Grar	Fabaceae	T	Impotency	R	DF	9	O	3	MA02
				Eye infection	B	DF	3	Op	2	
Vachellia tortilis (Forssk.) Galasso & Banfi	Korera	Fabaceae	T	Wound	L	F	4	De	4	MA04
Dichrostachys cinerea (L.) Wight & Arn.	Awrarise/Ader /Gorgoro	Fabaceae	S	Skin diseases	R	DF	2	De	2	MA40

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Vachellia oerfota (Forssk.) Kyal. & Boatwr.	Ajo, Chelegama	Fabaceae	T	Coughing	B	DF	5	O	1	MA01
				Scabies	L	F	3	De	2	
Calpurnia aurea (Ait.) Benth.	Digita	Fabaceae	S	Diarrhea	R	DF	5	O	12	MA16
				Snake bite	R	DF	5	O	2	
				Excessive bleeding after birth	R	DF	5	O	2	
Ocimum lamiifolium Hochst. ex. Benth.	Dama Kessie	Lamiaceae	H	Febrile disease	L	F	4	De	24	MA82
Ocimum basilicum L.	Besobila	Lamiaceae	H	Swelling	Wp	DF	4	De	1	MA81
				Stomachache	L	F	5	O	1	
Ocimum gratissimum (L.) subsp. *gratissimum*	Damakasse, Kesedama	Lamiaceae	S	Malaria	Wp	DF	4	De	3	MA83
Salvia nilotica Juss. ex Jacq.	Hulgeb	Lamiaceae	H	Wound	L	F	4	De	16	MA07
Rotheca myricoides (Hochst.) Steane & Mabb.	Misiroch	Lamiaceae	H	Wound	L	DF	4	De	3	MA32
Leonotis ocymifolia (Burml. f.) Iwarsson	Geram Tinjut, Eraskimir	Lamiaceae	S	Swelling	L	D	5	O	1	MA130
Ajuga integrifolia Buch.-Ham. Ex D. Don	Armagusa/Dem Akurt	Lamiaceae	H	Tonsillitis	L	F	5	O	1	MA129
Persea americana Mill.	Avocado	Lauraceae	T	Dandruff	Fr	F	4	De	2	MA88
Linum usitatissimum L.	Telba	Linaceae	H	Coughing	Se	D	7	Na	5	MA68
				Stomachache	Se	D	9	O	13	
Loranthella deflersii (Tiegh.) S.Blanco & C.E.Wetzel	Yebedena Tegedra	Loranthaceae	H	Dactylitis	L	F	3	De	1	MA85
Sida schimperiana Hochst. ex A. Rich.	Chifrig	Malvaceae	S	Eye disease	L	F	2	Op	2	MA108

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Malva verticillata L.	Lut/Adguar	Malvaceae	C	Vomiting	R	F	5	O	1	MA71
Stephania abyssinica (Quart.-Dill. & A.Rich.) Walp.	Yeayit Joro/Etse Eyesus	Menispermaceae	C	Pneumonia	L	F	5	O	2	MA115
Ficus sur Forssk.	Sholla	Moraceae	T	Skin diseases	LR	D	2	De	2	MA51
Ficus vasta Forssk.	Warka	Moraceae	T	Stomachache	R	F	5	O	1	MA52
Ficus carica L.	Beles	Moraceae	T	Wound	Lat	F	4	De	1	MA50
Moringa oleifera Lam.	Shiferaw/Mori nga	Moringaceae	T	Febrile disease	L	F	5	O	2	MA76
Myrsine africana L.	Kechemo	Primulaceae	S	Stomachache	LR	DF	14	O	3	MA77
Embelia schimperi Vatke	Enkoko	Primulaceae	S	Tap worm	Fr	D	13	O	1	MA43
Eucalyptus globulus Labill.	Nech Bahir Zaf	Myrtaceae	T	Common cold	L	F	5	Na	8	MA45
				Coughing	L	F	5	Na	6	
				Pneumonia	L	F	5	Na	2	
				Gout	L	F	10	O	1	
				Headache	L	F	5	Na	3	
				Febrile disease	L	F	10	Na	4	
Myrtus communis L.	Ades	Myrtaceae	S	Scabies	L	D	2	De	2	MA78
Psidium guajava L.	Zeyetun	Myrtaceae	T	Gastritis	Fr	F	9	O	4	MA93
Eucalyptus camaldulensis Dehnh.	Key Bahirzaf	Myrtaceae	T	Toothache	L	F	6	O	1	MA44
Jasminum abyssinicum Hochets. Ex DC.	Tenbelel	Oleaceae	C	Eye disease	L	F	2	Op	7	MA60
				Common cold	R	D	7	Na	10	
Olea europaea L. subsp. *cuspidata* (Wall. & G.Don) Cif.	Weira	Oleaceae	T	Dandruff	R	DF	2	De	12	MA84

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				Stomachache	L	F	5	O	7	
Olinia rochetiana A. Juss.	Tfie	Penaeaceae	S	Atopic eczema	L	D	3	De	1	MA131
Phytolacca dodecandra L'Hér.	Endod	Phytolaccaceae	S	Rabies	R	DF	5	O	3	MA90
				Malaria	R	F	5	O	4	
Saccharum officinarum L.	Shenkora Ageda	Poaceae	T	Coughing	SB	F	9	O	6	MA106s
Cymbopogon martini (Roxb.) Will.Watson	Teje Sar	Poaceae	H	Eye infection	L	F	3	Op	4	MA38
Hordeum vulgare L.	Gebs	Poaceae	H	Weight gain	SB	D	5	O	1	MA58
Afrocarpus falcatus (Thunb.) C.N.Page	Zigba	Podocarpaceae	T	Asthma	R	D	5	O	3	MA91
				Evil spirit	R	D	7	Na	2	
				Bone fracture	R	F	3	De	4	
				Abdominal pain	B	D	5	O	2	
Rumex nervosus Vahl	Embacho	Polygonaceae	S	Warts	L	DF	12	De	3	MA104
				Wound	L	DF	3	De	13	
				Fire burn	R	DF	3	Na	4	
Rumex nepalensis Spreng.	Kitel Rejim; Tult	Polygonaceae	H	Over blood flow after birth	L	F	17	NULL	2	MA103
				Dandruff	R	F	2	De	3	
				Stomachache	R	F	5	O	1	
				Wound	L	F	3	De	1	
Rumex abyssinicus Jacq.	Mekmeko	Polygonaceae	H	Intestinal parasite	R	D	15	O	2	MA102
Thalictrum rhynchocarpum Quart.-Dill. & A.Rich.	Sire-Bizu	Ranunculaceae	H	Stomachache	R	F	5	O	1	MA117
Clematis simensis Fresen.	Nech Yeazo-hareg	Ranunculaceae	C	Swelling	L	F	5	O	2	MA30

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Nigella sativa L.	Tikur Azmud	Ranunculaceae	H	Stomachache	Se	D	5	O	3	MA80
Ziziphus spina-christi (L.) Desf.	Kurkura, Geba	Rhamnaceae	T	Gonorrhea	R	DF	13	O	4	MA128
				Dandruff	L	F	4	De	2	
Ziziphus mauritiana Lam.	Kurkura	Rhamnaceae	T	Breast cancer	L	F	5	O	2	MA127
Rhamnus prinoides L'Hér.	Gesho	Rhamnaceae	S	Tonsillitis	L	F	11	O	2	MA95
Rosa abyssinica R.Br. ex Lindl.	Kega	Rosaceae	S	Tap worm	Fr	F	9	O	2	MA99
Prunus africana (Hook.f.) Kalkman	Tiqur Inchet	Rosaceae	T	Urinary disorders	SB	D	5	O	2	MA92
Rubus fruticosus L.	Enjori	Rosaceae	S	Gastritis	L	D	5	O	3	MA101
Rubia cordifolia L.	Mencherer	Rubiaceae	H	Coughing	L	DF	15	O	3	MA100
				Bone fracture	L	F	3	De	1	
				Stomachache	L	F	5	O	2	
Coffea arabica L.	Bunna	Rubiaceae	S	Asthma	Se	D	1	O	1	MA33
Citrus aurantiifolia (Christm.) Swingle	Lomi	Rutaceae	T	Minor bleeding	FrL	F	4	De	3	MA28
				Wound	Fr	F	4	De	5	
Ruta chalepensis L.	Tenadam	Rutaceae	H	Stomachache	Fr	F	5	O	4	MA105
				Evil Eyes	L	F	5	O	3	
				Common cold	L	F	5	O	3	
				Malaria	L	DF	1	O	4	
Citrus medica L.	Tiringo	Rutaceae	T	Appetite	Fr	DF	5	O	3	MA29
Osyris lanceolata Hochst. & Steud.	Keret	Santalaceae	S	Rabies	R	F	14	O	2	MA87
				Wound	L	F	2	De	1	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
Dodonaea viscosa subsp. *angustifolia* (L .f.) J.G.West	Kitkita	Sapindaceae	S	Atopic eczema	L	D	2	De	7	MA41
				Wound	L	DF	2	De	5	
				Eye disease/trachoma	R	D	2	Op	8	
Verbascum sinaiticum Benth.	Yahya Jero	Scrophulariaceae	H	Febrile disease	L	F	7	Na	11	MA120
				Wound	L	F	2	De	12	
Physalis lagascae Roem. & Schult.	Awut	Solanaceae	H	Varicella zoster virus	L	F	3	De	2	MA89
				Wound	L	F	3	De	5	
Nicotiana tabacum L.	Tinbaho	Solanaceae	S	Evil spirit	L	F	8	Na	3	MA79
				Common cold	L	F	4	Na	4	
Solanum anguivi Lam.	Zirch-embuay	Solanaceae	H	Toothache	R	D	15	O	4	MA110
Solanum dasyphyllum Schumach. & Thonn.	Geber-embuay	Solanaceae	S	Coughing	Fr	D	5	O	1	MA135
				Snake bite	R	F	9	O	1	
Solanum marginatum L.f.	Embuay	Solanaceae	S	Head injury (wound)	L	F	4	De	1	MA113
Capsicum annuum L.	Karia	Solanaceae	H	Malaria	Fr	F	9	O	1	MA19
Datura stramonium L.	Astenagir/Atse faris	Solanaceae	S	Dandruff	L	DF	4	De	1	MA39
				Toothache	Se	D	7	De	1	
Solanum somalense Franchet.	Yeshehochu Kitel /Shejerete Jin	Solanaceae	S	Headache	L	DF	7	Na	2	MA114
				Evil Eyes	L	DF	7	Na	5	
				Febrile disease	L	F	7	Na	2	

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				Fever	L	F	7	Na	3	
				Evil spirit	L	DF	7	O	3	
				Swelling	L	DF	3	De	1	
				Toothache	L	D	5	O	2	
				Diarrhea	L	DF	5	O	21	
Solanum incanum L.	Embuay	Solanaceae	S	Wound	L	DF	3	De	2	MA112
				Bleeding	L	F	9	O	4	
				Swelling	R	F	3	De	2	
				Stomachache	R	F	11	O	2	
				Bleeding	L	F	7	Na	2	
Capsicum frutescens L.	Mitmita Karia	Solanaceae	H	Flu	Fr	D	5	Na	5	MA20
Solanum lycopersicum L.	Timatim	Solanaceae	H	Malaria	L	F	11	O	3	MA70
Withania somnifera (L.) Dunal	Ede-buda/Gizawa	Solanaceae	S	Headache	R	DF	7	Na	3	MA124
				Evil Eyes	R	DF	7	Na	2	
				Gonorrhea	R	F	1	O	2	
				Syphilis	R	F	1	O	2	
				Febrile disease	L	F	7	Na	2	
				Fever	R	F	7	Na	1	
				Evil spirit	L	DF	7	O	2	
				Swelling	R	DF	3	De	3	
				Toothache	R	D	5	O	9	
Grewia ferruginea Hochst. ex A.Rich.	Lenkuata	Malvaceae	S	Stomachache	B	DF	11	O	2	MA55

Scientific names	Local name (Amharic)	Family name	GF	AT	PPU	CPU	MPAP	RA	UC	VS number
				Asthma	R	DF	11	O	3	
Grewia villosa Willd.	Agobday	Malvaceae	S	Broken bone	R	DF	3	De	1	MA56
Grewia bicolor Juss.	Sefa	Malvaceae	S	Stomachache	L	F	9	O	1	MA54
Celtis africana Burm.f.	Awrarise/Ameleka	Cannabaceae	T	Dactylitis	L	DF	3	De	1	MA25
Urtica simensis Hochst. ex A.Rich.	Sama	Urticaceae	H	Warts	L	F	4	De	4	MA119
Verbena officinalis L.	Atuch	Verbenaceae	H	Stomachache	R	DF	11	O	3	MA121
Lantana trifolia L.	Yeregna Kolo	Verbenaceae	S	Eye infection	Fr	D	4	De	1	MA66
Lippia abyssinica (Otto & A.Dietr.) Cufod.	Kessie	Verbenaceae	S	Stomachache	L	F	9	O	2	MA69
Lantana camara L.	Yewef Kolo	Verbenaceae	S	Chronic skin diseases	Fr	F	2	De	1	MA65
Tapinanthus globifer (A.Rich.) Tiegh.	Yebuna Tegedra	Loranthaceae	H	Diarrhea	L	F	5	O	4	MA123
Cyphostemma adenocaule (Steud. ex A.Rich.) Desc. ex Wild & R.B.Drumm.	Aserkush Tebetebkush	Vitaceae	C	Syphilis	L	F	2	De	2	MA24
				Herpes zoster	L	F	4	De	1	
Cissus quadrangularis L.	Yezhon Anjet	Vitaceae	C	Swelling	Wp	F	3	Na	2	MA27
Zingiber officinale Roscoe	Zingibil	Zingiberaceae	H	Stomachache	Rh	DF	1	O	7	MA126
				Common cold	Rh	DF	1	O	15	

Key: **Growth form, GF** (Tree, T; Shrub, S; Herb, H; Climber, C). **Ailment treated, AT**; **Plant part used**, **PPU** (Leaves, L; Root, R; Fruit, Fr, Seed, Se; Flower, Fl; Stem bark, SB; Latex, Lat; Bark, B; Bulb, Bu; Rhizome, Rh; The whole plant part, Wp; Hemiparasite, Hp; Leaves and Root, LR). **Conditions of part used, CPU** (Dry, D; Fresh, F; Dry and Fresh, DF). **Methods of preparation and application, MPAP** (1. Boil and drink the decoction when cool; 2. Grind and paint the powder or crushed part; 3. Grind, paste the crushed part and tie; 4. Extract the juice/oil/latex and pour or paint it; 5. Crush, homogenize with cold water and drink; 6. Hold with teeth; 7. Crush, heat/ burn or boil the part and inhale its smoke or steam; 8. Crush and sniff the freshly crushed part; 9. Eat the part; 10. Boil and do steam bath; 11. Drink the concoction; 12. Boil the part and paint the decoction; 13. Mixing the plant in local beverage, (Tella); 14. Mixing the plant in milk; 15. Mixing the plant with honey; 16. Mixing the plant with coffee; 17. Cut and drop on the ground).
Route of Administration, RA (Oral, O; Dermal, De; Nasal, Na; Optical, Op፡ Auricular, Au; Anal, An; Oral, Dermal, Nasal, ODN). **Use citation, UC**; **Voucher Specimen, VS**

Table 3.3: List of top 15 medicinal plants used for the treatment of human ailments in Habru District based on the use citation report by informants

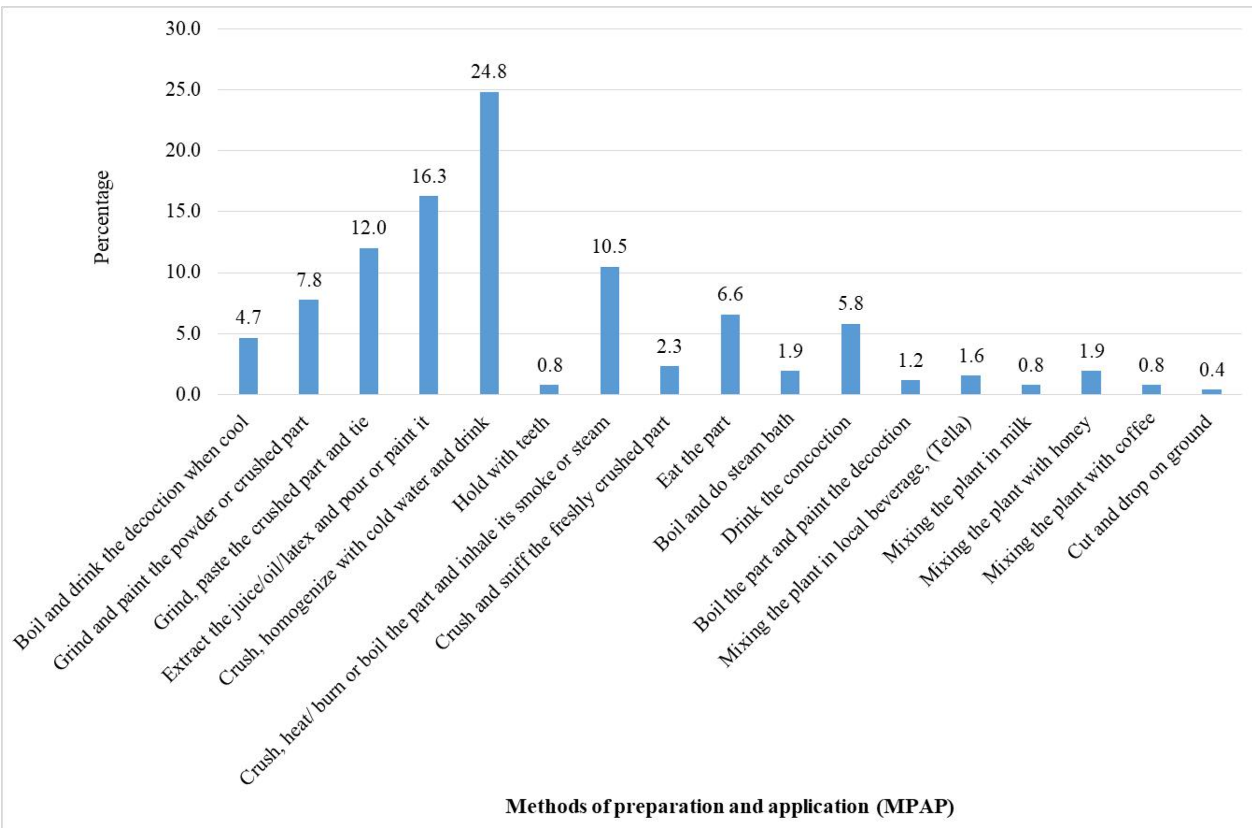
Scientific name	Use citation	Ailment treated in the study area	Ethnomedicinal use report to treat human ailments elsewhere in Ethiopia
Solanum somalense Franchet.	39	Headache, evil eyes, febrile disease, fever, evil spirit, swelling, toothache, diarrhea	Typhoid and fire burn (Tilahun Teklehaymanot, 2017)
Ehretia cymosa Thonn.	28	Evil spirit	Headache and abdominal pain (Getachew Ashagrie et al., 2023)
Cleome gynandra L.	27	Wound, ear infection, evil spirit	Diarrhea (Bizuneh Woldeab et al., 2018)
Croton macrostachyus Hochst. ex Delile	26	Atopic eczema, liver problem, stomachache, gonorrhea, malaria, chronic skin diseases, scabies, wound, minor bleeding, febrile disease	Atopic eczema (Abebe Ayele et al., 2022), Liver problem, stomachache (Getnet Chekole et al., 2015; Solomon Araya et al., 2015), gonorrhea (Balcha Abera, 2014; Getnet Chekole et al., 2015; Getnet Chekole, 2017), malaria (Balcha Abera, 2014; Solomon Araya et al., 2015), wound (Ermias Lulekal et al., 2013; Balcha Abera, 2014), minor bleeding (Ermias Lulekal et al., 2013), scabies (Ermias Lulekal et al., 2013; Solomon Araya et al., 2015; Getnet Chekole, 2017), tape worm (Getnet Chekole et al., 2015), gastritis (Banchiamlak Nigussie and Kim, 2019), bone fracture (Banchiamlak Nigussie and Kim, 2019), snake poison (Getnet Chekole et al., 2015), wound cancer (Nigatu Tuasha et al., 2018)
Withania somnifera (L.) Dunal	26	Headache, evil eyes, gonorrhea, syphilis, febrile disease, fever, evil spirit, swelling, toothache	Asthma/coughing (Getnet Chekole et al., 2015; Abebe Ayele et al., 2022), febrile disease (Abiyu Enyew et al., 2014; Getnet Chekole, 2017), typhoid (Tilahun Teklehaymanot, 2017), fever (Abiyu Enyew et al., 2014), swelling (Getnet Chekole, 2017; Tilahun Teklehaymanot, 2017), cancer (Nigatu Tuasha et al., 2018), malaria (Balcha Abera, 2014)
Eucalyptus globulus Labill.	24	Common cold, coughing, pneumonia, gout, headache, febrile disease	Common cold (Yayesh Limenih et al., 2015; Getnet Chekole, 2017; Banchiamlak Nigussie and Kim, 2019; Moa Megersa and Samuel Woldetsadik, 2022), coughing (Solomon Araya et al., 2015; Banchiamlak Nigussie and Kim, 2019; Abebe Ayele et al., 2022; Moa Megersa and Samuel Woldetsadik, 2022), headache (Banchiamlak Nigussie and Kim, 2019), febrile disease (Gidey Yirga, 2010; Getnet Chekole, 2017; Banchiamlak Nigussie and Kim, 2019; Yimer Assen et al., 2021; Moa Megersa and Samuel Woldetsadik, 2022), influenza (Balcha Abera, 2014)
Ocimum lamiifolium Hochst. ex. Benth.	24	Febrile disease	Febrile disease (Mirutse Giday et al., 2010; Solomon Araya et al., 2015; Yimer Assen et al., 2021; Moa Megersa and Samuel

Scientific name	Use citation	Ailment treated in the study area	Ethnomedicinal use report to treat human ailments elsewhere in Ethiopia
			Woldetsadik, 2022), ear infection (Mirutse Giday et al., 2010), coughing (Abebe Ayele et al., 2022), headache (Balcha Abera, 2014), parasites (Zewdie Kassa et al., 2020), diarrhea (Abiyu Enyew et al., 2014)
Verbascum sinaiticum Benth.	23	Febrile disease, wound	Febrile disease (Getnet Chekole, 2017), stomachache (Getnet Chekole et al., 2015; Getnet Chekole, 2017), snake bite (Abiyu Enyew et al., 2014), tonsillitis (Abebe Ayele et al., 2022)
Zingiber officinale Roscoe	22	Stomachache, common cold	Coughing (Abebe Ayele et al., 2022; Moa Megersa and Samuel Woldetsadik, 2022), stomachache (Gidey Yirga, 2010; Yayesh Limenih et al., 2015; Getnet Chekole, 2017; Yimer Assen et al., 2021), headache (Mirutse Giday et al., 2010), common cold (Moa Megersa and Samuel Woldetsadik, 2022), influenza (Balcha Abera, 2014), cancer (Nigatu Tuasha et al., 2018), malaria (Mirutse Giday et al., 2010), swelling (Getnet Chekole, 2017), tonsillitis (Abebe Ayele et al., 2022)
Allium sativum L.	21	Atopic eczema, asthma, common cold, dandruff, pneumonia, coughing, malaria	Skin infection (Tilahun Teklehaymanot, 2017), febrile disease (Alemayehu Kefalew et al., 2015; Getnet Chekole et al., 2015; Banchiamlak Nigussie and Kim, 2019), malaria (Abiyu Enyew et al., 2014; Balcha Abera, 2014; Getnet Chekole et al., 2015; Solomon Araya et al., 2015; Getnet Chekole, 2017; Banchiamlak Nigussie and Kim, 2019; Moa Megersa and Samuel Woldetsadik, 2022), intestinal parasite (Abiyu Enyew et al., 2014), stomachache (Getnet Chekole, 2017; Banchiamlak Nigussie and Kim, 2019), influenza (Mirutse Giday et al., 2003; Getnet Chekole et al., 2015; Moa Megersa and Samuel Woldetsadik, 2022), common cold (Moa Megersa and Samuel Woldetsadik, 2022), headache (Mirutse Giday et al., 2003, 2010), pneumonia (Getnet Chekole et al., 2015), coughing (Moa Megersa and Samuel Woldetsadik, 2022), wound (Solomon Araya et al., 2015)
Dodonaea viscosa subsp. *angustifolia* (L.f.) J.G.West	20	Atopic eczema, wound, eye disease/trachoma	Diarrhea (Banchiamlak Nigussie and Kim, 2019), eye disease/trachoma (Getnet Chekole, 2017), wound (Alemayehu Kefalew et al., 2015; Solomon Araya et al., 2015; Getnet Chekole, 2017), tape worm (Getnet Chekole et al., 2015)
Rumex nervosus Vahl	20	Warts, wound, fire burn	Stomachache (Alemayehu Kefalew et al., 2015), diarrhea (Yayesh Limenih et al., 2015), scabies (Abiyu Enyew et al., 2014), wound (Getnet Chekole, 2017), snake bite (Solomon Araya et al., 2015), breast cancer (Solomon Araya et al., 2015; Nigatu Tuasha et al., 2018), warts (Getnet Chekole et al., 2015). vomiting (Yayesh Limenih et al.,

Scientific name	Use citation	Ailment treated in the study area	Ethnomedicinal use report to treat human ailments elsewhere in Ethiopia
			2015)
Euclea racemosa L.	19	Wound	Dandruff (Getnet Chekole, 2017)
Olea europaea L. subsp. *cuspidata* (Wall. & G.Don) Cif.	19	Dandruff, stomachache	Asthma (Solomon Araya et al., 2015), psychiatric disease (Abiyu Enyew et al., 2014), tonsillitis (Abebe Ayele et al., 2022; Moa Megersa and Samuel Woldetsadik, 2022), gout (Alemayehu Kefalew et al., 2015), dandruff (Getnet Chekole, 2017), fever (Moa Megersa and Samuel Woldetsadik, 2022), tumor (Nigatu Tuasha et al., 2018), eye infection (Solomon Araya et al., 2015; Yayesh Limenih et al., 2015), snake bite (Tilahun Teklehaymanot, 2017)
Linum usitatissimum L.	18	Coughing, stomachache	Asthma (Banchiamlak Nigussie and Kim, 2019), liver disease (Banchiamlak Nigussie and Kim, 2019), wound (Getnet Chekole et al., 2015; Yayesh Limenih et al., 2015), placental retention (Solomon Araya et al., 2015), swelling (Gidey Yirga, 2010)

4.3.2. Disease types, modes of remedy preparation and application

Approximately 50 human health problems were reported in Habru District for which the local people reported being treated with medicinal plants. Wounds, stomachache, and diarrhea were the most commonly reported health problems (30% of the total human ailment reports) under the gastro-intestinal disease category, whereas atopic eczema, dandruff, swelling and warts were most frequently reported under the dermatological disease group. The traditional names and clinical explanations of the top ten most cited health problems are indicated in Table 3.4. The major modes of remedy preparation list were crush, homogenize with cold water and drink (24.8%), extract the juice/oil/latex and pour or paint it (16.3%), grind, paste the crushed part and tie (12%), and crush, heat/burn or boil the part and inhale its smoke or steam (10.5%) (Figure



3.5).

Figure 3.5: Methods of preparation and application of MPs in the study area

4.3.3. Plant parts and conditions used

Plant parts used for remedy preparation indicated that, leaves 122 (47.3%) were the widely used plant parts followed by roots 57 (22.1%), fruits 18 (7.0%) and seeds 15 (5.8%) (Figure 3.6). The local people of the study area reported employing plant materials of different conditions. The Fresh plant materials (parts) were the dominant ones accounting for 58% used in remedy preparation whereas both dried and fresh parts were used at least (24%); the remaining 17% remedies were reported to be prepared from dried parts of medicinal plant species.

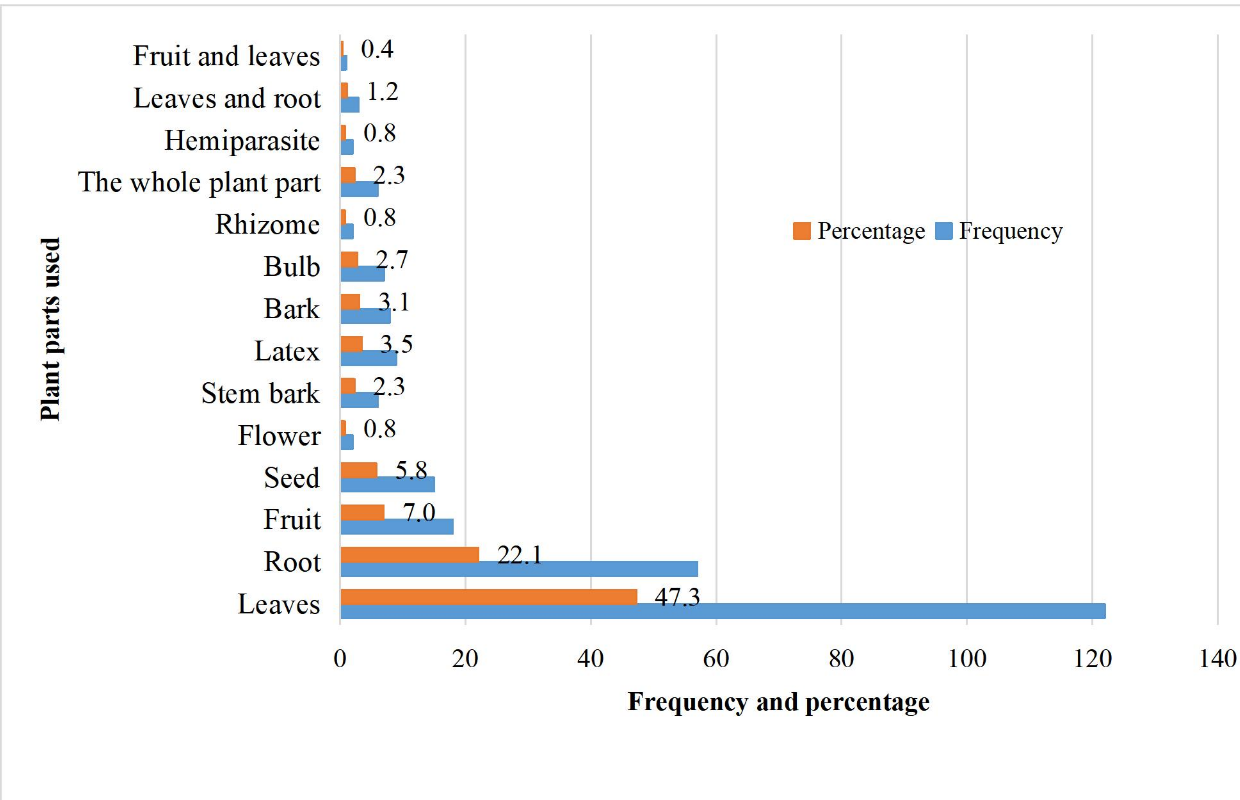


Figure 3.6: Plant parts used for remedy preparation in Habru District.

4.3.4. Routes of administration

In Habru district, different methods were used to administer medicinal plant preparations. Accordingly, the major routes of administration in the study area were reported to be oral application (122 preparations, 47.3%) followed by topical or dermal application (82 preparations, 31.8%). The detailes about routes of administration techniques are presented in Figure 3.7.

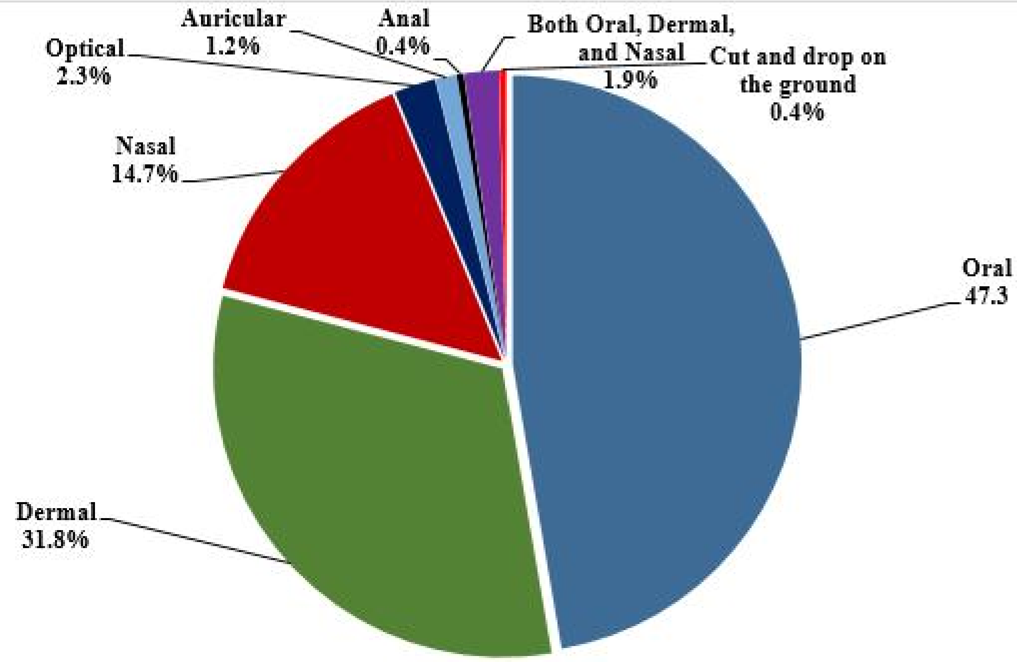


Figure 3.7: Routes of administration of traditional medicinal plants used in the study area

4.3.5. Marketability of medicinal plants

Among the reported medicinal plants in the study area, 22 (16.4%) species were reported as marketable and only six species (*Terminalia brownii* Fresen., *Myrtus communis* L., *Ruta chalepensis* L., *Olea europaea* L. subsp. *cuspidate* (Wall. & G.Don) Cif., *Allium sativum* L., and *Capsicum annuum* L.) were found in the local markets sold and purchased entirely for their medicinal applications (Figure 3.8). The majority of reportedly marketable medicinal plants (82%) were mainly sold for their non-medicinal uses but occasionally applied as medicine when the need arises. The average price of 25 – 40 cm long and 10 cm diameter *Terminalia brownii* Fresen. (WEYBA) stem at the Girana local market was 40 Birr (0.75 USD), whereas for a bunch (300-400 gm) of the branch material of *Myrtus communis* L. (ADES) was 10 Birr (0.21 USD); and the price was 15 Birr (0.28 USD) for a bunch (100 – 150 gm) of *Ruta chalepensis* L. (TENADAM) branch and fruit. A coffee cup of eight types of medicinal plant powder was sold for 15 - 20 Birr (0.28 – 0.37 USD) at Mehal Amba local market for treating dandruff, in which the seller was not interested in mentioning the name of these plants.

Figure 3.8: A) *Terminalia brownii* Fresen. at Girana kebele local market; B) *Allium sativum* L. and C) *Capsicum annuum* L.and D) *Myrtus communis* L. at Mehal Amba kebele local market; E) *Ruta chalepensis* L., F) *Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif., and G) a bunch of parts of eight processed medicinal plant parts said to treat problems related to wound at Mehal Amba kebele local market.

(Photo courtesy: Mulugeta Alemu, Habru District, Ethiopia, 2023).

Table 3.4: List of the top 10 most cited human health problems in the study area and their clinical descriptions

Local term in Amharic	Clinical term	Clinical descriptions
KUSIL	Wound	A disruption to the integrity of biological tissue, including skin, mucous membranes, and organ tissues, caused by various types of trauma (Kujath and Michelsen, 2008).
YEHOD KURTET	Stomachache	Abdominal pain and/or discomfort can arise from stomach-related issues such as parasites, infections, or allergies (Spangler et al., 2014).
YEMENFES BESHITA	Evil spirits	There is no clinical term for "evil spirit". The concept of evil spirits is a religious or spiritual one, not a medical one. In clinical terms, any symptoms or experiences that a person attributes to an evil spirit would be more likely explained by a mental health condition, such as a psychotic disorder, a dissociative disorder, or a seizure disorder (WHO, 2022).
MECHI/TIKUSAT	Febrile disease	It is characterized by the presence of fever, which is defined as an elevated body temperature beyond the normal range, usually caused by an infection and resulting from a higher body temperature set point (Raab et al., 2022).
GUNFAN	Common cold	It is an acute, self-limited viral infection of the upper respiratory tract, which may also involve the lower respiratory tract (Eccles, 2005).
TEKMAT	Diarrhea	Frequent passage of abnormally soft, liquid feces, a symptom of intestinal tract infection caused by a range of bacterial, viral, and parasitic organisms
SAL	Coughing	A reflex action that clears the throat and airways of foreign particles, mucus, or other irritants (Chung and Pavord, 2008).
YEAYN HIMEM	Eye Disease/trachoma	A chronic inflammatory disease of the eye and the leading cause of blindness (Evans et al., 2019).
FOREFOR	Dandruff	A scalp condition characterized by the presence of white or grayish flakes of dead skin cells, especially on the scalp (Ranganathan and Mukhopadhyay, 2010).
WEBA	Malaria	Malaria is a serious and sometimes fatal disease caused by parasites of the Plasmodium group and transmitted to people through the bites of infected female Anopheles mosquitoes (Walter and John, 2022).

4.3.6. Efficacy of medicinal plants

From the entirety of 50 distinct human ailments reported within Habru District, nine disease categories were identified (Table 3.5). Among these categories, those exhibiting the highest informant consensus factor (ICF) values were gastro-intestinal and parasitic ailments (0.85), followed closely by febrile diseases (0.84), cultural-related conditions such as evil spirits and evil eyes (0.84), and throat and respiratory diseases (0.80).

Table 3.5: ICF values of traditional medicinal plants for treating human ailments in Habru district

No.	Disease category (DC)	Reported diseases	Species	% of all species	Use citation	% of all use citation	ICF value
1	Gastro-intestinal and parasitic	Stomachache disease, tap worm, typhoid, giardia, gastritis, diarrhea, abdominal pain, constipation, intestinal parasite, vomiting	23	18.0	151	19.48	**0.85**
2	Dermatological	Atopic eczema, dandruff, scabies, wounds, chronic skin diseases, over blood flow after birth, excessive bleeding after birth, fire burn, minor bleeding, warts, swelling, varicella zoster virus, taking out the spine, boils	46	35.9	220	28.39	0.79
3	Throat and Respiratory Diseases	Tonsillitis, asthma, Common cold, Coughing, pneumonia	24	18.8	117	15.10	0.80
4	Febrile disease	Febrile disease, fever	11	8.6	63	8.13	**0.84**
5	Animals and insects cause poisonous diseases	Rabies, snake bite, Malaria	17	13.3	36	4.65	0.54
6	Evil spirits	Evil spirits, evil eyes	14	10.9	80	10.32	**0.84**
7	Organ diseases	Toothache, ear infection, ear mites, eye disease/trachoma, liver problems, jaundices	20	15.6	65	8.39	0.70
8	Musculoskeletal (Bone fracture	Bone fracture	3	2.3	6	0.77	0.60
9	Others	Syphilis, gingivitis, gout, headache, hemorrhoid, impotency, gonorrhea	15	11.7	44	5.68	0.67

4.3.7. Relative healing potential of medicinal plants

The highest fidelity level (91.3%) was recorded for *S. somalense* followed by *O. lamiifolium* (88.9%) and *V. sinaiticum* (85.7%) (Table 3.6). The recorded highest fidelity level values of *S. somalense* and *O. lamiifolium* were obtained under the gastro-intestinal and parasitic and febrile therapeutic categories respectively.

Table 3.6: Fidelity level value of ten medicinal plants commonly reported for use against a given ailment category

No.	Medicinal plant species	Therapeutic category	Np	N	FL value (%)	Rank
1	*Solanum somalense* Franchet.	Diarrhea	21	23	91.3	1
2	*Ocimum lamiifolium* Hochst. ex. Benth.	Febrile disease	24	27	88.9	2
3	*Verbascum sinaiticum* Benth.	Wound	12	14	85.7	3
4	*Withania somnifera* (L.) Dunal	Toothache	9	11	81.8	4
5	*Calpurnia aurea* (Ait.) Benth.	Diarrhea	12	16	75.0	5
6	*Rumex nervosus* Vahl	Wound	13	18	72.2	6
7	*Linum usitatissimum* L.	Stomachache	13	19	68.4	7
8	*Zingiber officinale* Roscoe	Common cold	15	22	68.2	8
9	*Euclea racemosa* L.	Wound	19	28	67.9	9
10	*Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif.	Dandruff	12	19	63.2	10

Where, FL= Fidelity Level, Np = number of informants who independently cited the importance of a species for treating a particular disease; N = total number of informants who reported the plant for any given disease

4.3.8. Use diversity of medicinal plants

The results obtained from the direct matrix ranking (DMR) exercise conducted on nine multipurpose medicinal plants enabled the identification of the specific plant facing the highest pressure within the area, along with the associated factors responsible for posing a threat to the plant. Accordingly, the DMR showed that, *O. europaea* subsp. *cuspidata* ranked first (most threatened) followed by *D. viscosa* subsp. *angustifolia* and *E. racemosa* (Table 3.7).

Table 3.7: Average DMR score of ten key informants for nine medicinal plant species with additional uses

Use diversity	Multipurpose medicinal plants subjected to DMR									Total	Rank
	Searsia retinorrhoea (Steud. ex Oliv.) Moffett	*Ehretia cymosa* Thonn.	*Juniperus procera* Hochst. ex Endl.	*Euclea racemosa* L.	*Croton macrostachyus* Hochst. ex Delile	*Eucalyptus globulus* Labill.	*Olea europaea* L. subsp. *Cuspidata* (Wall. & G.Don) Cif.	*Rumex nervosus* Vahl	*Dodonaea viscosa* subsp. *angustifolia* (L.f.) J.G.West		
A-Agricultural tools	5	4	2	3	2	2	4	0	4	26	4
B-House construction	4	3	5	5	3	5	5	1	5	36	**1**
C-Firewood	3	4	3	5	5	3	4	2	5	34	**2**
D-Fodder	0	1	0	1	0	0	0	3	1	6	6
E-Medicine	2	5	3	3	4	5	2	5	3	32	3
F-Charcoal	1	0	3	2	0	0	5	0	1	12	5
Total	15	17	16	19	14	15	20	11	19		
Rank	6	4	5	**2**	8	6	**1**	9	**2**		

Based on use criteria (5= excellent; 4= very good; 3=good; 2= less used; 1=least used and 0= no value)

4.3.9. Use of wild medicinal plants as awild edibles in the study area

In the study area, some plants have dual roles being used both as medicinal and wild edible resources. From the total reported medicinal plants, 11 species (8.2%) were identified as being utilized as wild edible plants in the study area. Among these plants, shrubs constituted 55%, while trees accounted for the remaining 45% in terms of growth habit. The edible plant parts utilized were diverse, with 82% of them being fruits, 9% comprising the inner parts of stems, leaves, and tender shoots, and 9% involving leaves, stems, and bark. In addition, the method of preparation varied with 82% of the plants featuring ripe fruit was consumed raw (Table 3.8). An additional 9% encompassed young tender shoots, leaves, and the inner part of stems, which are consumed fresh and raw. A distinct 9% of plants were found to be employed for flavoring traditional alcoholic beverages such as Tela (made from honey) and Tej (made from fermented grains).

Table 3.8: List of medicinal plants used as wild edible plants in Habru District

Scientific names	Local Name	Family names	Growth habit	Edible plant part	Method of preparation
Carissa spinarum L.	Agam	Apocynaceae	Shrub	Fruit	Ripe fruit eaten raw
Cordia africana Lam.	Wanza	Boraginaceae	Tree	Fruit	Ripe fruit eaten raw
Opuntia ficus-indica (L.) Mill.	Beles, Qulqual	Cactaceae	Shrub	Fruit	Ripe fruit eaten raw
Euclea racemosa L.	Dedeho	Ebenaceae	Shrub	Fruit	Ripe fruit eaten raw
Ficus sur Forssk.	Sholla	Moraceae	Tree	Fruit	Ripe fruit eaten raw
Ficus vasta Forssk.	Warka	Moraceae	Tree	Fruit	Ripe fruit eaten raw
Rumex nervosus Vahl	Embacho	Polygonaceae	Perenial herb	Inner part of stem, leaves and tender shoots	Young tender shoots, leaves and inner part of stem eaten fresh & raw
Ziziphus spina-christi Desf.	Kurkura, Geba	Rhamnaceae	Tree	Fruit	Ripe fruit eaten raw
Rhamnus prinoides L'Hér.	Gesho	Rhamnaceae	Shrub	Leave, Stem and bark	to flavor traditional alcoholic drinks (Tela and Tej)
Rosa abyssinica R.Br. ex Lindl.	Kega	Rosaceae	Shrub	Fruit	Ripe fruit eaten raw
Citrus aurantiifolia (Christm.) Swingle	Lomi	Rutaceae	Tree	Fruit	Ripe fruit eaten raw

4.3.10. Preference ranking

A preference ranking exercise with 15 key informants on medicinal plants that were reported to be used against diarrheal diseases, the most frequently reported disease in the gastro-intestinal and parasitic disease category, showed that *S. somalense* (yeshehochu kitel), *C. aurea* and *V. sinaiticum* (yeahiya joro) were the most preferred species to treat the reported disease (Table 3.9).

Table 3.9: Results of preference ranking of seven medicinal plants reported for treating diarrheal diseases

Medicinal plants reported for treating diarrheal diseases	Local Name in Amharic	Family name	Informants labelled KI1 to KI15															Total Score	Rank
			KI1	KI2	KI3	KI4	KI5	KI6	KI7	KI8	KI9	KI10	KI11	KI12	KI13	KI14	KI15		
Barleria eranthemoides	Yeset Af	Acanthaceae	1	3	3	1	2	1	2	1	4	5	3	3	4	8	7	**48**	**8**
Justicia schimperiana	Sensel	Acanthaceae	2	2	5	3	1	4	4	2	5	2	6	1	2	7	1	**47**	**9**
Carissa spinarum	Agam	Apocynaceae	4	1	1	5	6	6	1	4	3	1	2	6	3	2	5	**50**	**7**
Calpurnia aurea	Digita	Fabaceae	9	10	7	9	8	10	9	10	9	8	5	10	10	9	10	**133**	**2**
Olea europapaea L. subsp. *cuspidata*	Weira	Oleaceae	3	5	2	6	4	2	5	3	1	4	1	2	1	3	2	**44**	**10**
Ruta chalepensis	Tenadam	Rutaceae	6	8	4	4	5	3	3	7	6	3	7	5	8	5	3	**77**	**5**
Verbascum sinaiticum	Yahya Jero	Scrophulariaceae	7	6	9	8	10	7	8	8	8	9	8	7	6	4	8	**113**	**3**
Withania somnifera	Ede-buda/Gizawa	Solanaceae	8	7	8	7	7	8	7	6	7	7	10	9	5	6	6	**108**	**4**
Solanum somalense	Yeshehochu Kitel	Solanaceae	10	9	10	10	9	9	10	9	10	10	9	8	9	10	9	**141**	**1**
Rumex nervosus	Embacho	Polygonaceae	5	4	6	2	3	5	6	5	2	6	4	4	7	1	4	**64**	**6**

NB: Scores in the table indicate ranks given to medicinal plants based on their efficacy. The most important in the set is given the highest number, decreasing in number as the members of the set decrease in importance. This implies that, the highest number (10) given for the medicinal plant which informants thought most effective in treating evil spirits and the lowest number (1) for the least effective plant.

4.3.11. Traditional herbal medicine: opportunities, challenges, and indigenous conservation practices in Habru District

The participants in the FGD identified several opportunities for the utilization, regulation, and promotion of traditional medicine in the district. They emphasized the importance of raising awareness about traditional medicine (TM) and forest management practices. Additionally, the participants highlighted the need to strengthen research and development activities to further enhance the utilization of TM. They also expressed the potential for scaling up TM utilization by providing support through research-based inputs. Furthermore, the participants recognized the value of supporting traditional healers through training and material assistance. Continuous supervision from the Ministry of Health and other responsible organizations was seen as an opportunity to ensure the effective implementation of the study.

The FGD highlighted various challenges and threats that impact the utilization, regulation, and promotion of traditional medicine in the kebele. Among these, participants highlighted the detrimental effects of deforestation, agricultural expansion, firewood collection, and environmental degradation on medicinal plants. According to the responses from key informants, these factors collectively pose significant threats to the availability and sustainability of medicinal plants in the study area. The loss of habitat due to deforestation and agricultural activities, coupled with the increasing demand for firewood, has emerged as a prominent concern, potentially leading to the depletion of essential plant species used in traditional medicine. Furthermore, environmental degradation further exacerbates these threats, underscoring the urgent need for conservation efforts to safeguard valuable medicinal plant resources.

4.3.12. Indigenous and local knowledge on conservation practices

In this study, about 91.7% of the FGDs (11 out of 12) mentioned the cultivation of medicinal plants within home gardens; all FGDs (12 out of 12) stressed the importance of guarding against deforestation and fire; about 91.7% of the FGDs (11 out of 12) emphasized the need to control the massive harvest of wild medicinal plants; and finally, 100% of the FGDs (12 out of 12) highlighted the practice of maintaining seedlings in nurseries, planting, and overall conservation efforts as vital methods for safeguarding medicinal plant species.

To address the identified challenges and leverage the opportunities, the FGD participants put forth several recommendations. They called for intensified awareness campaigns about traditional medicine and forest management practices. Strengthening research and development activities emerged as a key recommendation to enhance the effectiveness of traditional medicine utilization. The participants also recommended scaling up the utilization of traditional medicine through research-based inputs and by offering support to traditional healers through training and material assistance. The need for continuous supervision from the Ministry of Health and other responsible organizations was underscored to ensure proper regulation and promotion of traditional medicine practices. Additionally, the participants emphasized the importance of addressing the issue of invasive plant species by implementing measures to replace them with native plants, thereby contributing to the conservation of plant diversity and the facilitation of traditional medicine utilization in the study kebeles.

4.4. Discussion

The ethnobotanical studies conducted in Ethiopia have revealed a rich and diverse knowledge of medicinal plants among the country's various ethnolinguistic groups (Banchiamlak Nigussie and Kim, 2019; Amare Bitew *et al.*, 2022). In Ethiopia, approximately 800 plant species are traditionally used to treat various human and livestock ailments (Edwards, 2001; Tilahun Teklehaymanot, 2009). The results of the present study are consistent with these findings, as they document the use of a wide variety of medicinal plants to treat a wide range of ailments in the country. The findings of this study underline the diversity and use of reported medicinal plants in Habru District. The taxa documented in this study (134 species belonging to 110 genera and 54 families) reflect the depth and breadth of the medicinal plants both taxonomically and in their medicinal lore as well as the functional attributes of each species. The list of medicinal plant species identified through ethnobotanical surveys highlights the rich botanical heritage of the area. The diversity of medicinal plants documented in the Habru District aligns with the studies conducted in adjacent areas. In Tenta District, South Wollo, Ethiopia, a total of 121 medicinal plant species were identified, with Fabaceae dominating the list (Tibebu Tefera and Mesele Yihune, 2019) and the trend is comparable to the Habru District's botanical landscape. Similarly, the study conducted in Gubalafto District, which neighbors the study area, 135 traditional medicinal plant species were documented, with Asteraceae being notably abundant (Getnet Chekole, 2017), aligning with the Habru District's diversity. Moreover, in Yalo Woreda, Afar regional state, Ethiopia, 106 medicinal plants were reported, emphasizing the prevalence of Fabaceae (Tilahun Teklehaymanot, 2017), a finding consistent with the Habru District and surrounding areas. These similar findings across different regions illustrate the prevalence and importance of specific plant families, underscoring the collective richness of medicinal plant diversity in these geographically adjacent areas. For example, this study identified species such as *R. chalepensis*, *O. europaea* L. subsp. *cuspidata*, *A. sativum*, *C. annuum*, *O. lamiifolium*, and *V. sinaiticum*, aligning with findings from studies conducted in Gubalafto, Tenta, and Yalo Districts. Additionally, frequently cited medicinal plants were mentioned in the study area also found in another region of Ethiopia such as *S. somalense* (Tilahun Teklehaymanot, 2017), *E. cymosa* (Getachew Ashagrie *et al.*, 2023), *C. gynandra* (Bizuneh Woldeab *et al.*, 2018), *C. macrostachyus* (Balcha Abera, 2014; Getnet Chekole *et al.*, 2015; Getnet Chekole, 2017; Banchiamlak Nigussie and Kim, 2019) and *W. somnifera* (Abiyu Enyew *et al.*, 2014; Getnet Chekole *et al.*, 2015; Getnet Chekole, 2017; Tilahun Teklehaymanot, 2017; Nigatu Tuasha *et al.*, 2018; Abebe Ayele *et al.*, 2022) which was in line with study in the neighboring regions. This may be attributed to the similar ecological landscape and cultural attributes of the neighboring regions.

Moreover, several studies in Ethiopia have reported Solanaceae as the most dominant medicinal plant family (Alemayehu Kefalew *et al.,* 2015; Solomon Araya *et al.*, 2015; Nigussie Amsalu *et al.*, 2018; Admasu Moges and Yohannes Moges, 2020; Netsanet Gonfa *et al.*, 2020; Birhanu Adibaru and Samuel Chane, 2021; Mikiyas Abebe, 2022; Moa Megersa and Samuel Woldetsadik, 2022). These studies showed that members of the Solanaceae family are renowned for their medicinal attributes and contain many phytochemicals that offer potential health benefits. Notable phytochemicals within this family encompass glycoalkaloids, anthocyanins, alkaloids, flavonoids, and terpenoids, as extensively documented (Elizalde-Romero *et al.*, 2021; Alagar Yadav and Sara Koshi, 2022). One study conducted in Seharti Samre District, Southern Tigray, Ethiopia found Solanaceae, Lamiaceae, and Fabaceae as the highest contributors of medicinal plants (Solomon Araya *et al.*, 2015). Another study conducted in Damot Woyde District, Wolaita Zone, Southern Ethiopia, found that Solanaceae was represented by 5 species, making it the third most common family of medicinal plants (Moa Megersa and Samuel Woldetsadik, 2022). Similarly, a study conducted in Ada'a District, East Shewa Zone of Oromia Regional State, Ethiopia, found Solanaceae to be one of the leading plant families that encompass large medicinal species (Alemayehu Kefalew *et al.*, 2015). The use of medicinal plants in the study area revealed a notable distribution, with the majority of species categorized as herbs (36%), followed by shrubs (32%) and trees (25%). This prevalence of herbs could be attributed to their proximity and ease of accessibility in nearby areas compared to trees and shrubs which are often harvested from more remote patches of forested areas. This finding agrees with the pattern of dominance of herbaceous species both in Ethiopia and other countries (Mirutse Giday *et al.*, 2003; Tabuti *et al.*, 2003; Haile Yineger *et al.*, 2007; Ermias Lulekal *et al.*, 2013).

Ethiopia is home to several endemic plant species, some of which are used for medicinal purposes. A review of Ethiopian endemic plants identified a total of 44 endemic medicinal plant species belonging to 20 families and 30 genera (Vivero *et al.*, 2005; Hiwot Ayalew *et al.*, 2022). In this study, the identification of two endemic medicinal plant species (*M. ferruginea* and *U. simensis*) in Habru District further emphasizes the unique ecological and botanical characteristics of the study district. These endemic species hold immense value in terms of their potential therapeutic properties and may contribute to the broader field of ethnopharmacology. The presence of such endemic medicinal plants signifies the distinctiveness of the local flora and its role in traditional healing practices.

The disease types identified, along with the diverse modes of remedy preparation and application, reflect the intricate traditional knowledge of the local community. Results revealed that gastrointestinal diseases and dermatological diseases are common health problems in the study area. Similar results were reported that, constipation and diarrhea are some of the most reported health problems under the gastrointestinal disease category in Ankober District, North Shewa Zone, Amhara Region, Ethiopia (Ermias Lulekal *et al.*, 2013). Whereas Fitsum Mardu *et al.* (2019) reported that, intestinal parasitic infections are common among prison inmates in Ethiopia. Traditional healers in the study area stated that they prepare remedies using a variety of methods, depending on the type of illness and the corresponding explanations by their patients. The utilization of different plant parts, often specific to certain conditions, showcases the careful selection and application of plant resources based on their perceived effectiveness. According to several studies on Ethiopian medicinal plants (Ketema Tolossa *et al.*, 2013; Balcha Abera, 2014; Admasu Moges and Yohannes Moges, 2020; Zewdie Kassa *et al*., 2020; Hiwot Ayalew *et al.*, 2022), leaves are the most used plant parts for remedy preparation, followed by roots and other plant parts such as seeds, stems, bark, fruits, young shoots, and flowers. In this study also leaves (122, 47.3%) cases are the most widely used plant part followed by roots. This is because these plant parts play a vital role in the whole life cycle of the plant species as they are the sites of various metabolic reactions and centers where high concentrations of secondary metabolites are found.

In the present study, fresh plant parts were the dominant ones (58.2%) used in remedy preparation against various human ailments. The use of freshly harvested plant parts is believed to enhance the efficacy of the remedies, as they are considered to contain higher levels of active ingredients of fresh plant parts that could be lost on drying. Similar findings were reported by Mirutse Giday *et al.* (2010); Ermias Lulekal *et al.* (2013); Zewdie Kassa *et al.* (2020); Mersha Ashagre and Ermias Lulekal (2021).

Routes of administration represent another dimension of traditional medicine, with various methods employed to harness the therapeutic benefits of medicinal plants. The study reveals the multifaceted ways in which these plants are integrated into local healthcare practices, ranging from oral ingestion to external applications. This diversity in routes of administration contributes to the versatility of traditional medicine. Accordingly, the major routes of administration in the study area were reported to be oral application (47.3%). Similar findings were reported by Ermias Lulekal *et al.* (2013); Getnet Chekole (2017); Zewdie Kassa *et al.* (2020) stating that, the oral route is the most common route of administration for medicinal plant preparations in Ethiopia. It involves consuming the remedies orally, either in the form of powders, decoctions, or infusions.

The results from the market survey of medicinal plants indicated that, only 16.4% of species were reported as marketable and the remaining 82.8% had no marketability report since they were not available in the local market of the district during the time of the research. Accordingly, *T. brownii*, *M. communis*, *R. chalepensis*, *O. europaea* subsp. *cuspidata*, *A. sativum* L., and *C. annum* were found on the local markets sold and purchased entirely for their medicinal applications. This indicates a good market demand for these plants, potentially leading to important economic returns for local communities involved in their conservation, trade, and utilization of MPs.

The highest recorded ICF values (0.85 and 0.84) indicated the best agreement among informants on the use of medicinal plant species reported to be used for treating gastro-intestinal and parasitic ailments and febrile diseases respectively. The efficacy of medicinal plants, closely tied to traditional healing practices, holds cultural and practical significance. The recognition of certain plants as highly efficacious underscores the importance of preserving and further exploring traditional knowledge for future healthcare advancements. The highest ICF values are important to identify plants of particular interest in the search for bioactive compounds (Heinrich *et al.*, 1998). Accordingly, a total of 23 medicinal plants of Habru District (with high ICF values) for treating gastro-intestinal and parasitic diseases are under investigation for their pharmacological properties by our research theme.

Fidelity level is a measure of the consistency of a plant's use for a particular ailment or purpose across different cultures or regions (Heinrich *et al.*, 1998). The finding in this study of the highest FL values for *S. somalense* (91.3%) against gastro-intestinal and parasitic diseases and *O. lamiifolium* (88.9%) against febrile diseases; and *V. sinaiticum* (85.7%) against wound could be considered the relative healing potential of medicinal plants against the corresponding diseases and provide valuable insights into local perceptions of efficacy and potency. Plants with the highest fidelity level values could also be targeted for further phytochemical investigation to prove the bioactive components and conservation efforts, as they may have important medicinal or cultural significance (Tangjitman *et al.*, 2015). Accordingly, further activity testing experiments are being carried out on extracts of these species by our research group. These understanding shapes preference ranking and contributes to the prioritization of certain plant species in healthcare practices.

The output of a DMR exercise showed the highest ranks for *O. europaea* subsp. *cuspidata*, *D. angustifolia* and *E. racemosa,* which are the multipurpose plant species (Moa Megersa and Samuel Woldetsadik, 2022) of the area based on analysis of the information collected from key informants. This result indicates that these plants are exploited more for their non-medicinal uses (house construction and firewood) than for reported medicinal values. Overharvesting of multipurpose medicinal plant species for house construction and firewood was found to be the responsible factor aggravating the depletion of the species in the area. Thus, the study findings highlight the need for immediate complementary conservation measures to save the fast-eroding multipurpose medicinal plant species in the study area. Research results also reported that multipurpose medicinal plant species are often overexploited for purposes other than their medicinal value, such as fuel wood, charcoal production, construction materials, and lumbering (Haile Yineger *et al.*, 2007; Abraha Teklay *et al.*, 2013; Ermias Lulekal *et al.*, 2013; Yimer Assen *et al.*, 2021; Moa Megersa and Samuel Woldetsadik, 2022).

The interconnection between medicinal and edible plants has been recognized across various cultures and regions. The utilization of medicinal plants as wild edible plants presents an opportunity for communities to benefit from their nutraceutical role (Aparicio *et al.*, 2021). Similarly, a study in South Africa found that several wild edible vegetables were used for medicinal purposes (Mokganya and Tshisikhawe, 2019). Overall, the utilization of medicinal plants as wild edible plants in Ethiopia contributes to food security, provides nutritional benefits, and showcases the rich traditional knowledge of local communities. Further research is needed to explore the nutritional and medicinal properties of specific plant species and their potential for sustainable food systems (Haile Tesfaye, 2020; Muhidin Tahir *et al.*, 2021; Mulatu Hankiso *et al.*, 2023). The preference ranking exercise also helped to identify the most-preferred medicinal plant species to treat diarrheal diseases under the gastro-intestinal and parasitic disease categories. Accordingly, *S. somalense* (YESHEHOCHU KITEL), *C. aurea* (DIGITA.) and *V. sinaiticum* (YEAHIYA JORO) scored the highest values indicating that they are the most preferred species to treat diarrheal diseases in the study area.

Generally, this study of traditional medicinal plants in Habru District revealed the opportunities and challenges facing the plant and knowledge resources. The opportunities identified by the local community, including raising awareness, strengthening research and development activities, and supporting traditional healers, indicate a potential path toward the enhanced utilization and regulation of herbal medicine. However, challenges such as deforestation, agricultural expansion, and environmental degradation pose significant threats to medicinal plant resources, necessitating focused conservation efforts. The findings of this study indicated the intricate interplay among the biodiversity, cultural practices, and healthcare systems in Habru District. The diverse array of medicinal plants, along with their traditional applications, provides a valuable foundation for further exploration, conservation, and potential integration into modern healthcare practices.

According to the comparison of our findings with other ethnobotanical study in Ethiopia, novel plant uses of some medicinal plants were documented. *S. somalense* was used to treat headache, febrile disease, fever, swelling, toothache, and diarrhea, whereas *C. gynandra* for treating wound and ear infection was completely novel use in the study area and never ever reported. In general, the documented medicinal plant species and associated knowledge have the potential to contribute for the future public health initiatives and the development of sustainable herbal medicine practices in Habru District. The findings of this study can be used to inform the development of culturally sensitive public health interventions, empower local communities to manage their own health needs, and guide future research on the therapeutic properties of the identified plants.

4.5. Conclusion

This study indicated the relationship between the local community and the diverse array of medicinal plant species in Habru District. Traditional medicinal plant species are potential sources in the primary traditional healthcare systems of the people in the study area. The identification of 134 plant species, including 2 endemics, underscores the area's unique biodiversity and its role in conserving useful plants. The participation of informants from diverse demographics has enriched our understanding, revealing variations in ethnobotanical knowledge across gender, age groups, literacy levels, and marital status. The study relied on triangulated data collection and analysis techniques considering both emic and etic perspectives and using both qualitative and quantitative approaches. Such approaches enhance the validity of our findings and reveals of the depth of the community's practices and the key plant species that capture researchers' attentions. The documentation of MPs in terms of use value (UV), PR scores, and FL values would empower the potential to strengthen future pharmaceutical and phytochemical explorations, as well as conservation initiatives. Consequently, it becomes imperative to focus on safeguarding the traditional medicinal plants and the associated indigenous and local knowledge within the study area and beyond, ensuring their sustainable use and continuity into the future. These findings serve as valuable resources for sustainable conservation strategies, healthcare practices, and the preservation of traditional knowledge, underscoring the intricate interdependence of human societies and their natural surroundings.

Traditional healers and the local people who use medicinal plants in Habru District would need support from the education sector, tailored training, and finance to gain better knowledge of medicinal plant conservation and improve the mode of sustainable utilization. Further research needs to be conducted on antimicrobial, antioxidant and phytochemical profiling of potentially effective medicinal plants used in the study area, priority being given to *S. somalense*, *V. sinaiticum*, *R. nervosus, W. somnifera* and *C. aurea,* all of which are used against diarrheal diseases, found to be common in the study area and elsewhere in Ethiopia and beyond to be used as an input for future pharmacological research and development.

CHAPTER FIVE

5. Antibacterial Activity and Phytochemical Screening of Traditional Medicinal Plants Most Preferred for Treating Infectious Diseases in Habru District, North Wollo Zone, Amhara Region, Ethiopia

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Antibacterial Activity and Phytochemical Screening of Traditional Medicinal Plants Most Preferred for Treating Infectious Diseases in Habru District, North Wollo Zone, Amhara Region, Ethiopia

Abstract

Ethiopia's healthcare system relies on traditional medicinal practices that use medicinal plants to treat human and livestock ailments. However, the lack of empirical validation regarding the efficacy of these treatments against specific infectious diseases necessitates rigorous scientific investigations. The objective of this study was to investigate the antibacterial activity and phytochemical screening on five selected medicinal plant species, namely *Solanum somalense* Franchet., *Verbascum sinaiticum* Benth., *Rumex nervosus* Vahl, *Withania somnifera* (L.) Dunal and *Calpurnia aurea* (Ait.) Benth. The plants were first identified jointly with local informants and later considering mainly their high informant consensus and fidelity level values for their efficacy in treating infectious diseases in the area. Ethanol and aqueous extracts were prepared from the plant materials, and their antibacterial activities were evaluated against standard bacterial strains, representing both gram-positive and gram-negative types. To assess the antibacterial activity of the extracts, the minimum inhibitory concentration (MIC) was determined using the broth dilution method. Additionally, phytochemical screening was performed using standard qualitative tests to identify various secondary metabolites. The results indicated antibacterial efficacy in the ethanol extracts of *S. somalense, W. somnifera,* and *C. aurea* against particular bacterial strains (*S. somalense* against *S. agalactiae* with MIC of 1.5 mg/mL; *W. somnifera* against *S. aureus* and *E. coli*, with MIC of 2 mg/mL; *C. aurea* against *E. coli* and *K. pneumoniae*, with MICs of 2 mg/mL and 3 mg/mL, respectively). The results of the phytochemical screening indicated the presence of steroids, alkaloids, flavonoids, saponins, and terpenoids. The selected medicinal plants demonstrated promising antibacterial activity against certain bacterial strains. The current findings support the long-standing claim of the traditional medical system of the study area for their continued use of these plants in their treatment of infectious diseases. Further investigation is required to isolate the responsible active compounds and characterize the constituents and description of their antibacterial effect for possible use in areas where these infectious diseases are major health problems.

Keywords: Antibacterial, Traditional Medicine, Phytochemical, Plants, MIC

5.1. Introduction

Despite significant advancements in modern medicine, microbial diseases persist as substantial global threats (Cos *et al.*, 2006). Acknowledging this, the World Health Organization emphasizes herbal medicine as a primary healthcare modality in numerous developing nations (WHO, 2001). Globally, approximately 80% of the population relies on medicinal plants for disease treatment, with a more pronounced prevalence in African nations (WHO, 2001). Ethiopia, recognized for its rich biodiversity and profound traditional knowledge of medicinal plants, significantly incorporates these resources into its healthcare system. Recent research conducted by Nigatu Tuasha *et al.* (2018); Admasu Moges and Yohannes Moges (2020) reaffirms that around 80% of the population and 90% of livestock in Ethiopia depend on traditional medicine, attributing medicinal plants as the cornerstone of these practices and emphasizing their role in treating various ailments, including bruised ulcer, malaria, diabetes, cancer, and infectious diseases.

Plants are vital resources for drug discovery due to their extensive historical use in traditional medicine (Fabricant and Farnsworth, 2001). The Food and Agriculture Organization report indicates that at least 25% of pharmaceutical drugs in modern pharmacopoeia are plant-derived, with numerous others being synthetic analogues derived from plant prototype compounds (Food and Agriculture Organization (FAO), 2000). Throughout history, natural products from plants have served as medicine (Cowan, 1999; Bolla *et al.*, 2011) displaying a diverse array of bioactive compounds effective against various infectious agents (Chitravadivu *et al.*, 2009). Vital methods for assessing bioactive properties and identifying compounds from plant extracts include antibacterial activity testing and phytochemical screening (Edeoga *et al.*, 2005; Ahmad and Aqil, 2007).

Previous research has explored the antibacterial and phytochemical properties of *Verbascum sinaiticum* Benth. (Scrophulariaceae), *Rumex nervosus* Vahl (Polygonaceae), *Withania somnifera* (L.) Dunal (Solanaceae) and *Calpurnia aurea* (Ait.) Benth. (Fabaceae) (Kalkidan Lulseged *et al.*, 2022; Minda Asefa *et al.*, 2022; Dessie Belay *et al.*, 2021; Hazman *et al.*, 2021; Gebremedhin Romha *et al.*, 2018; Shemsu Umer *et al.*, 2013; Sener and Dulger, 2009). However, there is no investigation report available concerning the antibacterial and phytochemical properties of *Solanum somalense* Franchet. (Solanaceae). Despite the existing research on these plants, there is still a need for further investigation into their antibacterial and phytochemical properties. In developing countries like Ethiopia, where access to modern drugs is constrained, the impact of microbial diseases is notably pronounced (Ermias Lulekal *et al.*, 2014). The Habru District, situated in the North Wollo Zone of the Amhara Regional State, mainly relies on traditional medicine within its primary healthcare system. Despite its significance, the botanical diversity and ethnobotanical practices in this region lack comprehensive study and documentation.

Consequently, this study aims to evaluate the antibacterial activity and phytochemical constituents of select medicinal plants, considering the degree of informants' consensus and fidelity level values for treating infectious diseases.

5.2. Materials and methods

5.2.1. Selection of medicinal plants and plant material collection and preparation

Ethnobotanical methods were applied as recommended by (Martin, 1995; Alexiades, 1996) and ethnobotanical data on herbal use and preparation of the medicinal plants were collected by interviewing general informants, herbalists, and key informants. The ethnobotanical field study was carried out from November 26/2022 – December 14/2022 in Habru District. This study encompassed a total of 388 informants (250 males and 138 females) from all 13 kebeles within Habru District. The selection of these informants followed systematic random and purposive sampling methods, including peer recommendations, as described by (Martin, 1995). Qualitative and quantitative ethnobotanical data were collected from informants through a pre-prepared semi-structured interview method (Martin, 1995; Alexiades, 1996; Cotton, 1996). In the broader context of this study, techniques such as group discussions, semi-structured interviews, guided field walks, market surveys, preference ranking, and direct matrix ranking were employed (Martin, 1995). Additionally, a total of 13 focus group discussions were carried out, one at the district level and 12 at selected kebeles of the district. In each focus group discussion key informants, traditional healers, elders, kebele and district administrative officials from natural resource and forest protection offices, and agricultural and rural development offices were involved to amplify insights into medicinal plant knowledge at the community level and to corroborate information obtained through semi-structured interviews. Along with collection of ethnobotanical data, voucher specimens of traditional medicinal plant species were collected, taxonomically identified, authenticated, confirmed by a taxonomic expert, and deposited at the National Herbarium of Ethiopia, AAU. From among the plants recorded, five species (Table 4.1) that were frequently recommended by traditional herbalists and knowledgeable elders for their efficacy in treating infectious diseases and have relatively higher informant consensus and fidelity level values were selected for antibacterial activity and phytochemical constituents (Fabricant and Farnsworth, 2001).

In a recent study, we identified *Solanum somalense*, *Verbascum sinaiticum*, *Rumex nervosus*, *Withania somnifera and Calpurnia aurea* as key medicinal plants within the Habru District, Amhara Region, Ethiopia (Mulugeta Alemu *et al.*, 2024). Building upon this foundational research, fresh leaves of these selected species were collected for further analysis. The collected plant material was dried at room temperature and using solar drier (35 – 40^0C), and the plant cutlets were milled to powder. The powder was accurately weighed using an electronic weighing balance, then packed into polyethylene bags to prevent air entry and contamination. Finally, the bags were stored in a securely closed container, appropriately labeled, to facilitate for further extraction processes.

Table 4.1: Ethnobotanical data on medicinal plants selected for antibacterial tests and phytochemical screening.

Scientific name	**Family name**	**Local name (Amharic)**	**Parts used**	**Locally used in the study area to treat**	**FL value (%)**	**Frequencies of citations**	**Voucher no.**
Solanum somalense Franchet.	Solanaceae	Yeshehochu Kitel /Shejerete Jin	Leaves	Headache, **diarrhoea**, evil eyes, febrile disease, fever, evil spirits, swelling, toothache	91.3	39	MA114
Verbascum sinaiticum Benth.	Scrophulariaceae	Yahya Jero	Leaves	Febrile disease, **wound**	85.7	23	MA120
Rumex nervosus Vahl	Polygonaceae	Embacho	Leaves, root	Warts, **wound**, fire burn diarrhoea	72.2	20	MA104
Withania somnifera (L.) Dunal	Solanaceae	Ede-buda/Gizawa	Leaves, root	Headache, **toothache**, evil eyes, syphilis, gonorrhoea, febrile disease, fever, evil spirits, swelling	81.8	26	MA124
Calpurnia aurea (Ait.) Benth.	Fabaceae	Digita	Leaves and root	**Diarrhoea**, snake bite, excessive bleeding after birth	75	16	MA16

Remark: The FL value % in the table represents the importance of each species in treating specific diseases, as indicated by the bold entries in the disease category column. A higher FL value signifies a greater significance of the respective species in the treatment of the mentioned ailments in the study area.

Preparation of extracts

The dried plant material of each species was finely ground and the powder was extracted by maceration while shaking using orbital shaker successively until exhaustion, 80% ethanol (500 mL) and aqueous/distilled water were used as extraction solvents (Kloucek *et al.*, 2005; Abdelah Bogdadi *et al.*, 2007). The 80% ethanol extracts from each plant were subsequently filtered using Whatman filter paper followed by concentration under reduced pressure (vacuum) by using a rotary evaporator (R-300 Buchi, Switzerland) *in vacuo* at 40^{o}C to give gummy residue while the aqueous extracts of each plant were lyophilized to yield of amorphous powder (Cos *et al.*, 2006; Abdelah Bogdadi *et al.*, 2007). Finally, the concentrated extracts were dried and kept in a refrigerator until used for the experiment.

Microorganisms

The antibacterial activity of leaf extracts of the selected MPs was evaluated against selected standard bacterial strains of the American Type Culture Collection (ATCC). The bacterial strains selected were representative of both classes of Gram-positive and Gram-negative bacteria. The tested gram-positive bacterial strains were *Staphylococcus aureus* (ATCC 25923), *Streptococcus agalactiae* (ATCC 12386), *Staphylococcus epidermidis* (ATCC 12228), *Enterococcus faecalis* (ATCC 29212) and those of the gram-negative bacterial strains included *Proteus mirabilis* (ATCC 35659), *Salmonella typhimurium* (ATCC 13311), *Klebsiella pneumoniae* (ATCC 700603), *Escherichia coli* (ATCC 25922) and *Shigella flexneri* (ATCC 12022). The microorganisms were maintained at the Traditional and Modern Medicine Research Directorate (TMMRD) of Armauer Hansen Research Institute (AHRI) microbiology laboratory on Triptosoya + 20% glycerol broth at -78°C. Finally, the selected bacterial strains were cultivated using Mueller-Hinton broth (Basiru et al., 2013).

Ethical consideration

Ethical considerations were carefully addressed throughout the study to ensure the protection of community knowledge and the rights of the participants. Official permission from relevant authorities and local stakeholders (Addis Ababa University, Department of Plant Biology and Biodiversity Management, Armauer Hansen Research Institute, and Habru District Administration) was obtained to conduct the research in the study area. Prior to the commencement of interviews for ethnobotanical data collection, informed consent was obtained from each participant. This process involved a thorough explanation of the study's objectives and an assurance of judicious use of the information gathered, in accordance with ethical research standards. Confidentiality and anonymity were maintained during data collection, analysis, and reporting to safeguard the privacy of the participants. Ethical guidelines and principles were strictly adhered to throughout the study to uphold the highest standards of ethical conduct in research. Researchers must adhere to all applicable institutional, national, and international regulations and guidelines when conducting experimental research or field studies on plants, including when collecting plant specimens.

5.2.2. Antibacterial assay

Determination of minimum inhibitory concentration

The antibacterial activity test to determine the minimum inhibitory concentration (MIC) of a crude 80% ethanol and aqueous extract were conducted using the 96-well microtiter plate broth dilution method. The MIC of the plant extracts was determined using the protocols established by the Clinical and Laboratory Standards Institute (CLSI) guidelines (CLSI, 2018; Eloff, 1998). To prepare the stock solutions of the plant extracts, each extract was dissolved in distilled water and 5% tween 80, resulting in a concentration of 32 mg/mL for each solution. Quadruplicate wells were prepared in the 96-well microplates for each bacterial strain, with 100µl of Muller Hinton Broth (MHB) added to each well. Then, 100µl of the extract was added on the first row and two-fold serial dilutions with multichannel micropipettes were made down from row A to row H. Thus, serial dilutions ranging from 16 mg/mL to 0.125 mg/mL were prepared using Mueller-Hinton broth in the 96-well microplates, using a stock solution as the source. To obtain the active culture, the stored stock culture was sub-cultured at 37°C overnight on Mueller Hinton Agar. A few bacterial colonies were transferred from the sub-cultured plate to tubes containing sterile MHB using a sterile disposable inoculating loop. The test organism was standardized by preparing a diluted inoculum with an optical density (OD) range of 0.08 to 0.1 at 625nm, equivalent to a bacterial concentration of 1.0×10^8 colony-forming units per milliliter (CFU/mL), following the guidelines provided by the CLSI, 2018. Test organism suspensions with a concentration of approximately 5×10^5 CFU/mL well were prepared by diluting the standardized inoculum in MHB. Within 15 minutes of standardization, 100 µl of the bacterial test suspensions (5×105 CFU/mL) were added to each well of the microtiter plate, except for the sterility control (column 12). The plate was then incubated at 37°C for 18-24 hours. After incubation, a 40 µl solution of 0.4 mg/mL 2,3,5-Triphenyltetrazolium chloride (TTC) was added to each well as an indicator of microbial growth and incubated at 37°C for 30 minutes. The MIC values were determined by visually observing the presence or absence of pink color using magnifying instruments after the 30-minute incubation period. The MIC was recorded as the lowest concentration of each extract that did not display visible pink color, indicating no microbial growth. The MIC values were determined by conducting quadruplicate measurements. For each strain, a sterility control well and a growth control well were included in the study. To assess the bacterial sensitivity, parallel experiments were carried out using ciprofloxacin as a positive control, starting at a concentration of 10 µg/mL in sterile water (range of 10 µg/mL to 0.078125 µg/mL). Additionally, a negative control experiment was conducted using distilled water and 5% tween 80.

Determination of minimum bactericidal concentration

The determination of the minimum bactericidal concentration (MBC) was conducted following established protocols (Eloff, 1998; Clinical and Laboratory Standards Institute, 2018). To determine the MBC, streaking was performed on media plates by taking 5µl liquid part from each MIC well plate that showed no growth. The sub-cultured (inoculated) plates were then incubated under suitable conditions at 37°C for 18-24 hours. The growth of bacteria corresponding to different extract concentrations was meticulously examined. The plant extract concentration at which bacterial growth on freshly inoculated agar plates totally inhibited was identified as the MBC. To ensure robust results, all experiments were conducted in triplicate (Kang *et al.*, 2011).

5.2.3. Phytochemical screening

Phytochemical screening was conducted on both ethanol and aqueous extracts using the methods outlined by Basiru *et al.* (2013); María *et al.* (2018); Shaikh and Patil (2020); Kilonzo and Munisi (2021) to determine the presence of secondary metabolites. Preliminary phytochemical screening of the *S. somalense*, *V. sinaiticum*, *R. nervosus*, *W. somnifera* and *C. aurea* leaf aqueous extracts were tested for steroid, alkaloid, flavonoid, saponin, tannin, phenol, cardiac glycosides, terpenoid, coumarin, anthraquinine and anthocyanins. The selection of these metabolites was based on their superior potency in terms of antibacterial activity. The results were denoted as (+) for the presence and (-) for the absence of phytochemicals.

Test for steroid

In the phytochemical screening for steroids, two tests were conducted. In the first test, known as the Salkowski test, 2 mL of chloroform and a concentration of H_2SO_4 were added to 5 mL of the aqueous plant extract. A positive result was indicated by the presence of a red color in the lower chloroform layer (Muhamad *et al.*, 2022). In the second test, a mixture was prepared by combining 5 ml of chloroform and 5 ml of H_2SO_4 with 500 µl of the plant extract. A positive test was observed when the color changed from violet to blue or green, or when a ring of blue-green appeared. Furthermore, if the upper layer exhibited a change from yellow to green fluorescence in the H_2SO_4 log, it also indicated a positive result (Basiru *et al.*, 2013).

Test for alkaloid

In the test for alkaloids, two tests were performed. In the first test, known as Dragendroff's test, approximately 1 mL of the extract was placed in a test tube, and a few drops of Dragendroff's reagent, a commonly used alkaloid detection reagent, were added carefully to the test tube (Firdouse and Alam, 2011; Basiru *et al.*, 2013; Shaikh and Patil, 2020). The presence of alkaloids was determined by the formation of a reddish-brown precipitate. In the second test, known as Wagner's test, 1 ml of the extract was treated with a few drops of Wagner's reagent, a solution of iodine in potassium iodide (Basiru *et al.*, 2013). Again, the presence of alkaloids was indicated by the occurrence of a reddish-brown precipitate.

Test for flavonoids

In the phytochemical screening for flavonoids, two tests were conducted. In the first test, known as the alkaline reagent test, 2 mL of the plant extract was treated with 5 drops of dilute sodium hydroxide (NaOH), followed by the addition of diluted hydrochloric acid (HCl). The presence of flavonoids was indicated by the observed color change during the test, specifically when the yellow solution treated with NaOH turned colorless upon the addition of dilute HCl (Onwukaeme *et al.*, 2007). In the second test, the plant extract sample was mixed with a 10% ammonium hydroxide solution in a test tube. A positive result for flavonoids was indicated by the presence of yellow fluorescence (Shaikh and Patil, 2020).

Test for saponins

To detect the presence of saponins, 3 mL of the extract was vigorously shaken with 3 ml of distilled water in a test tube (Basiru *et al.*, 2013; Hossain *et al.*, 2013; Bharadwaj, 2020). The formation of foam or froth upon shaking was used as an indicator of a positive result.

Test for tannin and phenol

In the test for tannin and phenol, 2 mL of $FeCl_3$ solution was added to 1 mL of the plant extract. A positive result is indicated by the presence of a black or blue-green color (Kancherla *et al.*, 2019).

Test for tannin

Two ml of the tested plant extract was stirred with 3 mL of distilled water, followed by the addition of five drops of ferric chloride (FeCl). A positive test for tannins was indicated by the formation of a dark blue precipitate (Kumar *et al.*, 2007; Bharadwaj, 2020).

Test for phenol

A few drops of dilute iodine solution were added to 1 mL of the plant extract. A positive test for phenols was indicated by the transient appearance of a red color (Tiwari *et al.*, 2011; Shaikh and Patil, 2020).

Test for cardiac glycocide

Keller-Killiani test was employed. A mixture was prepared by combining 1 ml of the plant extract, 1.5 mL of glacial acetic acid, and 1 drop of 5% $FeCl_3$, followed by the addition of a concentrated solution of H_2SO_4 along the side of the test tube. The presence of a blue color in the layer of the glacial acetic acid solution indicated a positive test for cardiac glycosides (Basiru *et al.*, 2013; Kancherla *et al.*, 2019).

Test for terpenoid

In the method for testing terpenoids, approximately 5 ml of the plant extract was added to a mixture of 3 mL of chloroform and 2 ml of concentrated sulfuric acid (H_2SO_4). A positive test for terpenoids was indicated by the observation of a grey or reddish-brown color (Edeoga *et al.*, 2005).

Test for coumarin

A NaOH test was performed by adding 10% NaOH and chloroform to the plant extract. A positive test for coumarin was indicated by the presence of a yellow color (Kumar *et al.*, 2018).

Test for anthraquinone

In the test for anthraquinone, the Borntrager's test was employed. A mixture was prepared by combining 10 mL of 10% ammonium solution with a few ml of the filtrate, followed by vigorous shaking for 30 seconds The observation of a pink violet or red color indicated a positive test result for anthraguinine (Njoku and Obi, 2009; Gul *et al.*, 2017; María *et al.*, 2018).

Test for anthocyanins

In the method for testing anthocyanins, the HCL test was conducted by combining 2 mL of the plant extract with 2 mL of 2N HCL, followed by the addition of a few ml of ammonia. A positive test for anthocyanins was indicated by the transformation of the pink-red solution into a blue-violet color upon the addition of ammonia (Obouayeba *et al.*, 2015; Shaikh and Patil, 2020).

5.3. Results

5.3.1. Antibacterial activity

The antibacterial activity of the five plant leaf extracts (S. *somalense, V. sinaiticum, R. nervosus, W. somnifera,* and *C. aurea*) evaluated against the selected bacterial strains showed different positive results. The MIC results in mg/mL indicate the lowest concentration of each plant extract at which visible growth of the bacteria was not observed. The antibacterial activity of extracts was screened against gram positive strains *(S. aureus, S. agalactiae*, *S. epidermidis*, *E. faecalis)* and gram-negative bacterial strains (*P. mirabilis*, *S. typhimurium*, *K. pneumoniae*, *E. coli* and *S. flexner* and results obtained are summarized in Table 4.2.

Table 4.2: Antibacterial activity of 80% ethanol and aqueous leaf extracts against pathogenic organisms

	Bacterial Strains	Plant extracts along with MIC result in mg/mL (Mean ± SD)										Cipro (µg/mL)	MBC
		S1		V2		R3		W4		C5			
		Ethanol	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous		
Gram-positive bacteria strains	*Staphylococcus aureus* (ATCC 25923)	5 ±2.0	12±4.62	4±0.0	16±0.0	8±0.0	16±0.0	2±0.0	2±0.0	10±4.0	16±0.0	0.625±0.0	>16
	Streptococcus agalactiae (ATCC 12386)	1.5±0.58	16±0.0	4±0.0	16±0.0	16±0.0	16±0.0	3±1.15	4±0.0	16±0.0	16±0.0	0.625±0.0	>16
	Staphylococcus epidermidis (ATCC 12228)	12±4.62	16±0.0	12±4.62	16±0.0	8±0.0	16±0.0	3±1.15	4±0.0	16±0.0	16±0.0	0.625±0.0	>16
	Enterococcus faecalis (ATCC 29212)	4±0.0	16±0.0	4±0.0	16±0.0	16±0.0	16±0.0	3.5±1.0	4±0.0	16±0.0	16±0.0	1.25±0.0	>16
Gram-negative bacteria strains	*Proteus mirabilis* (ATCC 35659)	6±2.31	16±0.0	7±2.0	16±0.0	16±0.0	16±0.0	16±0.0	16±0.0	8±0.0	16±0.0	0.078±0.0	>16
	Salmonella typhimurium (ATCC 13311)	4±0.0	16±0.0	4±0.0	8±0.0	16±0.0	8±0.0	8±0.0	8±0.0	10±4.0	16±0.0	0.469±0.0	>16
	Klebsiella pneumoniae (ATCC 700603)	3±1.15	16±0.0	5±2.0	12±4.62	8±0.0	12±4.6	8±0.0	16±0.0	3±1.15	12±4.62	0.469±0.0	>16
	Escherichia coli (ATCC 25922)	3.5±1.0	12±4.62	5±2.0	16±0.0	12±4.62	16±0.0	2±0.0	16±0.0	2±0.0	16±0.0	0.352±0.0	>16
	Shigella flexneri (ATCC 12022)	8±0.0	8±0.0	3±1.15	4±0.0	16±0.0	16±0.0	4±0.0	3±1.15	12±4.62	16±0.0	1.250±0.0	>16

Key: S1: *Solanum somalense* Franchet. (Solanaceae), V2: *Verbascum sinaiticum* Benth. (Scrophulariaceae), R3: *Rumex nervosus* Vahl (Polygonaceae), W4: *Withania somnifera* (L.) Dunal (Solanaceae), C5: *Calpurnia aurea* (Ait.) Benth. (Fabaceae), Cipro: Ciprofloxacin. The extracts were diluted in a series of steps, starting with a concentration of 16 mg/mL and ending with a concentration of 0.125 mg/mL. Ciprofloxacin was used as a positive control, starting at a concentration of 10 µg/mL and diluting down to 0.078125 µg/mL. The comprehensive raw data supporting this study can be found in the Supporting Information section (S1 File).

The ethanolic extract of *S. somalense* exhibited significant antibacterial activity against *S. agalactiae, K. pneumoniae*, and *E. coli*, with MICs of 1.5 mg/mL, 3 mg/mL, and 3.5 mg/mL, respectively. Similarly, the ethanolic extract of *W. somnifera* showed activity against *S. aureus* and *E. coli*, with an MIC of 2 mg/mL. Additionally, the ethanolic extract of *C. aurea* demonstrated activity against *E. coli* and *K. pneumoniae*, with MICs of 2 mg/mL and 3 mg/mL, respectively (Figure 4.1 and 4.2).

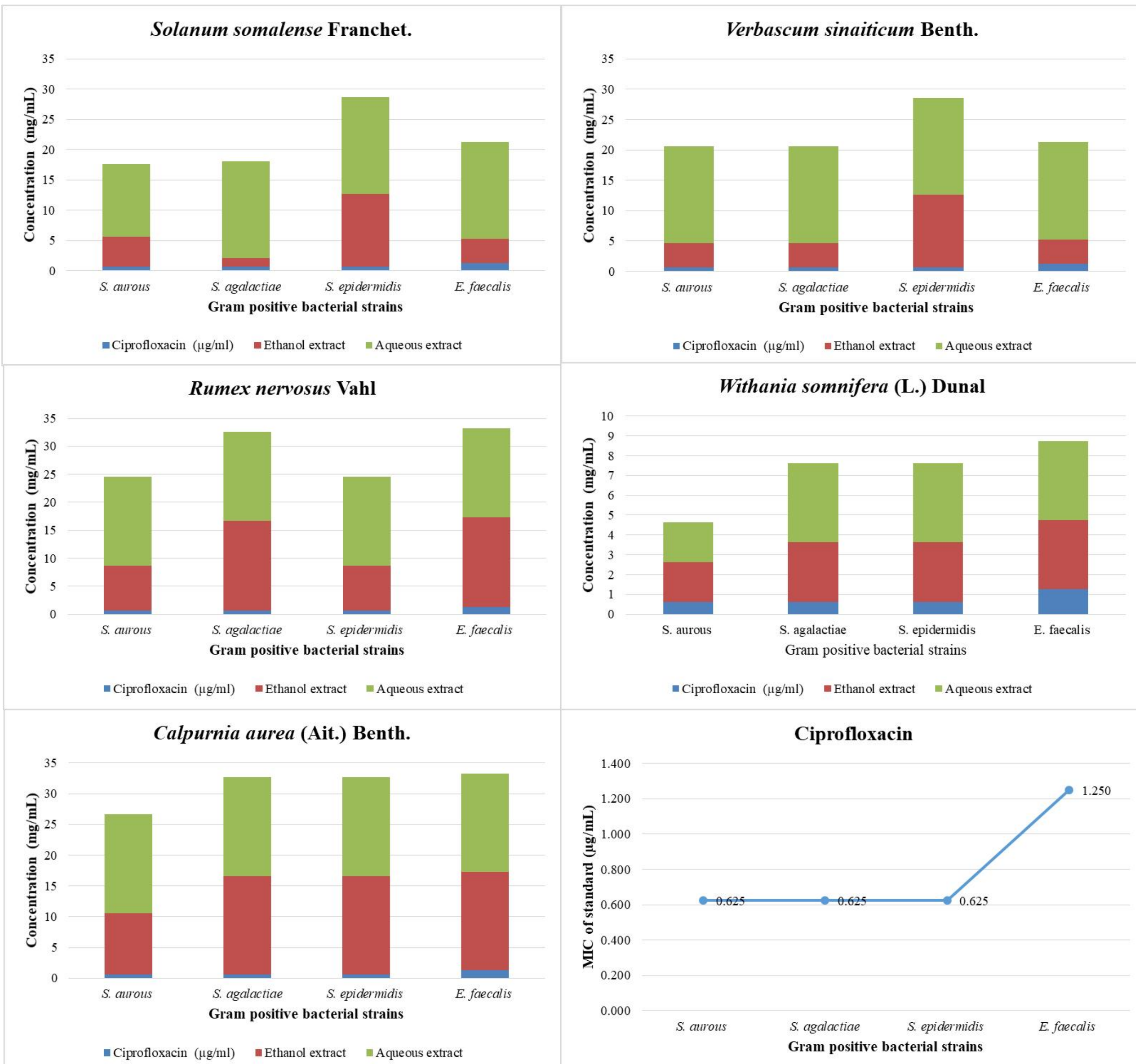

Figure 4.1: MIC of selected plant extracts against gram positive bacterial strains.

Ciprofloxacin, a commonly used antibiotic used as a positive control in the study, exhibited MIC values ranging from 0.078125 µg/mL to 1.25 µg/mL against the tested bacterial strains. This indicates the potency of ciprofloxacin in inhibiting bacterial growth at relatively low concentrations compared to the plant extracts (Figure 4.2).

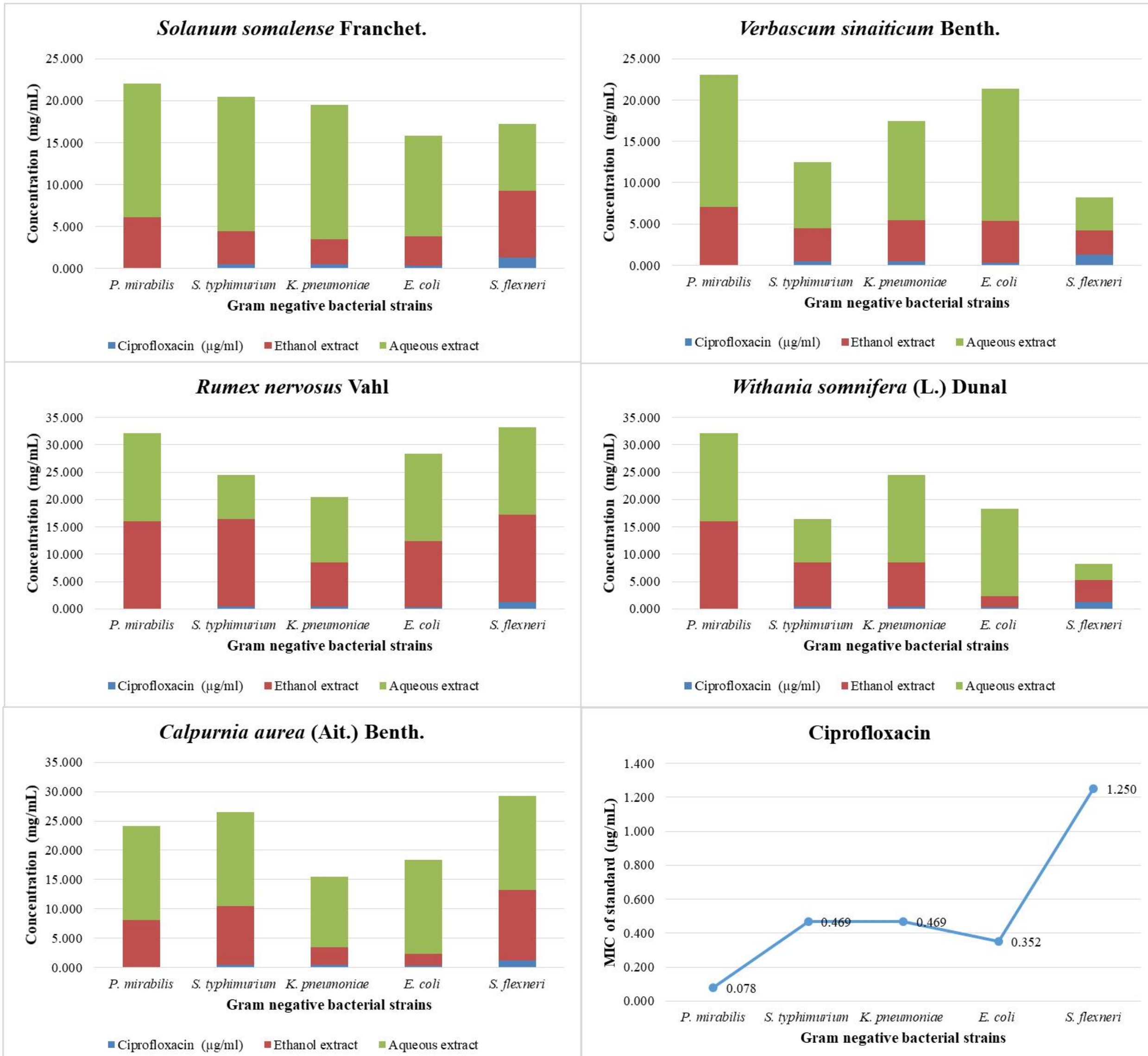

Figure 4.2: MIC of selected plant extracts against gram negative bacterial strains

For the gram-positive bacteria strains, the aqueous extract of *V. sinaiticum* exhibited lower MIC values compared to the ethanol extract. This suggests that the aqueous extract of V2 is more effective at MIC of 4 mg/mL in inhibiting the growth of *S. aureus, S. agalactiae,* and *E. faecalis*. On the other hand, the MIC values of the ethanol extract of *S. somalense, R. nervosus,* and *W. somnifera* were relatively higher, indicating less potent antibacterial activity against these Gram-positive bacteria strains. Ciprofloxacin, the positive control, displayed lower MIC values, indicating its strong inhibitory effect on these bacteria. Among the Gram-negative bacteria strains, the aqueous extract of *V. sinaiticum* again demonstrated lower MIC values compared to the ethanol extract. This suggests its stronger antibacterial activity against *P. mirabilis, S. typhimurium, K. pneumoniae, E. coli,* and *S. flexneri.*

The MBC values were determined to assess the bactericidal effect of the plant extracts. However, the results indicated that none of the tested plant extracts exhibited bactericidal activity at the MIC tested concentrations. This implies that the concentrations of the plant extracts that inhibited bacterial growth were not sufficient to completely kill the bacteria.

5.3.2. Phytochemical screening

Phytochemical screening of ethanolic extracts of five MPs (*S. somalense, V. sinaiticum*, *R. nervosus*, *W. somnifera*, and *C. aurea*) was carried out to identify either the presence or absence of secondary metabolites which are steroid, alkaloid, flavonoid, saponin, tannin, phenol, cardiac glycosides, terpenoid, coumarin, anthraguinine and anthocyanins as shown in Table 4.3. All tested extracts exhibited positive results for steroids, which involved the preparation of a mixture comprising 5 mL of chloroform, 5 mL of H_2SO_4, and 500 µl of the plant extract. A positive test was indicated by the formation of a blue-green ring. The phytochemical screening of *S. somalense* demonstrated positive results for steroids, alkaloids, flavonoids, saponins, and terpenoids. Tested extracts of *V. sinaiticum* and *C. aurea* exhibited the presence of the specified chemical compounds, except for anthocyanins. Notably, anthraquinones were detected only in *V. sinaiticum.* Tannins and phenolic compounds were found to be abundant in the tested samples, potentially in the form of tannins, as indicated by the formation of a dark precipitate upon treatment with a ferric chloride solution. However, *S. somalense* did not show the formation of a dark precipitate in the ferric chloride test. Cardiac glycosides, as detected by the Keller-Killiani test, and coumarins, as indicated by the NaOH test, were exclusively present in *V. sinaiticum* and *C. aurea*. Alkaloids, steroids, and flavonoids were found to be relatively common in the ethanolic extracts, indicating their presence in significant amounts. On the other hand, anthraquinones and anthocyanins were observed to be the least abundant compounds in the tested extracts.

Table 4.3: Phytochemical screening tested medicinal plants

Phytochemical Components	Test	Plant Extracts				
		S1	V2	R3	W4	C5
Steroids	Salkowski test	-	+	-	+	-
	Chloroform + H_2SO_4	+	+	+	+	+
Alkaloids	Dragendroff's test	-	-	-	-	-
	Wagner’s test	+	+	-	-	+
Flavonoids	Alkaline reagent test	+	+	+	-	+
	Alkaline reagent test using 10% ammonium hydroxide sol.	-	+	-	-	-
Saponin	Foam test	+	+	+	-	+
Tannins and Phenolic	$FeCl_3$ test	-	+	+	+	+
Tannins	Ferric chloride (FeCl) test	-	+	+	+	+
Phenolic	Iodine test	-	+	+	+	+
Cardiac Glycosides	Keller-Killiani test	-	+	-	-	+
Terpinoides	Salkowski test	+	+	+	-	+
Coumarins	NaOH test	-	+	-	-	+
Anthraquinones	Borntrager's test	-	+	-	-	-
Anthocyanins	HCL test	-	-	-	-	-

Key: + = positive; - = negative; S1: *S. somalense*, V2: *V. sinaiticum*, R3: *R. nervosus*, W4: *W. somnifera*, C5: *C. aurea*

5.4. Discussion

The results of the present study showed that the ethanolic extracts of the plants that had higher informant consensus and fidelity level values (*S. somalense, W. somnifera* and *C. aurea)* exhibited antibacterial activity against the selected bacterial strains. The ethanolic extract of *S. somalense* demonstrated antibacterial activity against *S. agalactiae, K. pneumoniae,* and *E. coli*, with MICs of 1.5 mg/mL, 3 mg/mL, and 3.5 mg/mL, respectively. This indicates the potential of *S. somalense* as a source of antibacterial compounds. The ethanolic extract of *W. somnifera* also showed activity against *S. aureus* and *E. coli*, with an MIC of 2 mg/mL. This suggests that *W. somnifera* has antibacterial properties against gram-positive and gram-negative bacteria.

Likewise, the MICs of 3 mg/mL and 3.5 mg/mL were recorded for the ethanolic extract of *C. aurea* against *E. coli* and *K. pneumoniae,* respectively. This indicates the potential of *C. aurea* as an antibacterial agent.

Numerous studies (Hailu Tadeg *et al.*, 2005; Shemsu Umer *et al.*, 2013; Dharajiya *et al.*, 2014; Ermias Lulekal *et al.*, 2014; Hazman *et al.*, 2021; Kalkidan Lulseged *et al.*, 2022; Minda Asefa *et al.*, 2022) conducted in various countries have reported a significant success rate in antibacterial activity tests, aligned with the ethnomedicinal background. A synthesis of findings from various studies reveals a consistent pattern of antibacterial efficacy across a diverse range of plants, despite differences in phytochemical composition and geographical origins. Notably, research by Ermias Lulekal *et al.* (2014) highlights that 74% of ethanol extracts from various plants exhibited antimicrobial activity, with *E. schimperi*, *O. lamiifolium*, and *R. steudneri* showing broad-spectrum activity. This aligns with the findings of Rawat and Bisht (2014), who reported the potent antibacterial activity of the methanolic extract of *W. somnifera* against gram-positive clinical isolates. Similarly, Dharajiya *et al.* (2014) found that methanol extracts of *W. somnifera* were highly effective against *E. coli* and *B. cereus*. Al Yahya *et al.* (2018) contributed to this body of knowledge by demonstrating the significant antimicrobial activity of methanol and n-hexane extracts of *R. nervosus*. Dessie Belay *et al.* (2021) extended this understanding by revealing the antimicrobial and antioxidant activities of solvent fractions of *C. aurea* leaves. Furthermore, Shemsu Umer et al. (2013) and Gebremedhin Romha et al. (2018) both observed promising antibacterial activities in methanol and chloroform extracts of *C. macrostachyus* Del., with *Calpurnia aurea* extracts exhibiting the highest zone of inhibition against *S. aureus*. Also, Solomon Berhanu (2022) found that extracts of *C. aurea* were effective against both *E. coli* and *S. aureus*, contributing to the growing evidence of the plant's antibacterial properties. These studies predominantly align with our research in terms of the ethnomedicinal plants examined, although there are variations in species across different regions. The significant success rates in antibacterial activity tests reported in these studies corroborate the ethnomedicinal use of these plants, reinforcing the validity of traditional knowledge in identifying potential antibacterial agents.

Overall, the results of the present study suggest that the ethanolic extracts of *S. somalense*, *W. somnifera*, and *C. aurea* have potential as antibacterial agents. The methanol extract of *W.*

somnifera stem showed the maximum antibacterial activity against *E. coli.* The range of MIC of extracts recorded was 3.125 to 50 mg/mL (Dharajiya *et al.*, 2014), with the lowest MIC value of 3.125 mg/mL being recorded for methanol extract against *E. coli*. The paper reported the presence of alkaloids, tannins, saponins, cardiac glycosides, steroids, phenols, and flavonoids in the methanol and aqueous extracts of the stem of *W. somnifera.* However, glycosides and sterol were absent in the hexane extract, and cardiac glycoside was absent in the ethyl acetate extract. Overall, the aqueous extracts of *V. sinaiticum* and *C. aurea* demonstrated better antibacterial activity against both gram-positive and gram-negative bacteria strains compared to the ethanol extracts. However, none of the tested plant extracts exhibited bactericidal effects at the tested MIC concentrations. This suggests that the extracts may inhibit the growth of bacteria but do not completely kill them.

Phytochemical screening of the ethanolic extracts of the five medicinal plants revealed the presence of various secondary metabolites. All tested extracts exhibited positive results for steroids in the Chloroform and H_2SO_4 test. *S. somalense* showed positive results for steroids, alkaloids, flavonoids, saponins, and terpenoids. This suggests that *S. somalense* contains a diverse range of bioactive compounds. *V. sinaiticum* and *C. aurea* exhibited the presence of most chemical compounds tested, except for anthraquinones, which were only detected in *V. sinaiticum.* Anthocyanins were not detected in any of the tested extracts. The absence of Anthocyanins in all the tested extracts may be attributed to several factors. Anthocyanins are water-soluble pigments responsible for the vibrant red, purple, or blue colors in various fruits, vegetables, and flowers (Lila, 2004). They are commonly found in plants belonging to the families Rosaceae, Solanaceae, and Ericaceae, among others, which include fruits like strawberries, blueberries, and cherries. *S. somalense* did not show the formation of a dark precipitate in the ferric chloride test, indicating a difference in the presence of tannins compared to other plants. Cardiac glycosides and coumarins were exclusively present in *V. sinaiticum* and *C. aurea*. Alkaloids, steroids, and flavonoids were relatively common in the ethanolic extracts, indicating their significant presence. Indeed, understanding the precise modes of action can provide insights into how these compounds interact with bacterial cells, potentially leading to the development of more effective antimicrobial agents.

The results obtained from various studies further support the potential of these plants as sources of antibacterial agents and highlight their diverse biological activities. Ethanol extracts obtained from the leaves of *V. sinuatum* L. exhibited strong antibacterial activity against *E. faecalis* and *P. mirabilis* (Sener and Dulger, 2009). This finding reinforces the potential of *V. sinuatum* L. as an effective antibacterial agent. Another species of Verbascum, *V. lasianthum* Boiss. ex Bentham, was investigated for its biological activity and element content by (Hazman *et al.*, 2021). The study revealed that different concentrations of *V. lasianthum* extracts possess antibacterial activity, along with antioxidant and cytotoxic properties. This further supports the medicinal potential of Verbascum species.

In addition to antibacterial activity, *V. sinaiticum* has also been explored for its wound healing and antioxidant properties. The hydroalcoholic leaf extract of *V. sinaiticum* in excision and incision wound models (Kalkidan Lulseged *et al.*, 2022). The results demonstrated significant wound healing efficacy, as evidenced by an enhanced wound contraction rate. This suggests that *V. sinaiticum* extracts have potential applications in wound healing therapies. Furthermore, *V. sinaiticum* has been found to possess analgesic and anti-inflammatory properties. 80% methanol root extract of *V. sinaiticum* exhibited peripheral and central analgesic effects, along with anti-inflammatory activity (Minda Asefa *et al.*, 2022). These beneficial effects could be attributed to the presence of phytochemicals such as alkaloids, tannins, flavonoids, phenols, steroids, and glycosides.

Regarding *C. aurea*, it has also demonstrated antimicrobial activity against various bacterial strains. The activity of *V. sinaiticum* and *C. aurea* against *S. aureus* and *E. coli*, with different minimum inhibitory concentrations (Hailu Tadeg *et al.*, 2005). This indicates the potential of *C. aurea* as an effective antimicrobial agent. The ethanol extract of *C. aurea* leaves and bark and found significant antimicrobial activity against *E. coli* and *S. aureus* (Gebremedhin Romha *et al.*, 2018; Solomon Berhanu, 2022). The study also identified the presence of various phytochemicals, including phenolic compounds, saponins, flavonoids, alkaloids, and others in the plant extracts. These compounds contribute to the antibacterial potential of *C. aurea*.

Moreover, the antimicrobial activity of the 80% methanol extract of *C. aurea* leaves against a range of organisms, including *Salmonella typhimurium, Shigella* species, *P. aeruginosa, S. aureus*, and *E. coli* (Shemsu Umer *et al.*, 2013). The methanol extract of *C. aurea* exhibited moderate inhibition of bacterial growth, with the highest activity observed against *E. coli*. Additionally, the antimicrobial effects of 95% methanol extracts of *C. aurea* leaves (Dessie Belay *et al.*, 2021). Their study revealed significant antimicrobial activity of the ethyl acetate and dichloromethane fractions of the leaves against pathogenic bacteria, including *S. aureus*, *E. coli*, *E. faecalis,* and *K. pneumoniae*. These findings provide further support for the potential of *C. aurea* as a valuable source of antibacterial compounds.

The positive outcomes observed in the antibacterial activity tests coupled with the presence of bioactive compounds in these extracts establish a promising foundation for pharmaceutical exploration. For instance, the correlation between the detected bioactive compounds, such as alkaloids, flavonoids, and steroids in *S. somalense* aligns with its observed antibacterial potential against specific bacterial strains, thereby validating its traditional use in ethnomedicine. Similarly, the presence of diverse chemical constituents, such as tannins and phenols, in *W. somnifera* and *C. aurea* echoes their noted antibacterial activity against different bacteria, corroborating their historical medicinal applications. While our study suggests the possibility of novel antimicrobial agent development from these plants, further investigations are crucial to decipher the precise mechanisms of action and potential synergies among these bioactive compounds. This comprehensive understanding bridges the traditional knowledge of these medicinal plants with our scientific findings and may pave the way for the development of novel antimicrobial agents with potential pharmacological benefits.

5.5. Conclusion

The current study demonstrated the antibacterial activity of the selected medicinal plant extracts against gram-positive and gram-negative bacterial strains. The MIC results highlighted the inhibitory concentrations of the extracts, while the MBC results indicate the absence of bactericidal effects at the tested concentrations. Accordingly, the extracts of *S. somalense, W. somnifera* and *C. aurea* exhibit varying degrees of antimicrobial activities at MIC range from 1.5 - 3.5 mg/mL against pathogenic microbes namely *S. aureus, S. agalactiae, K. pneumoniae* and *E. coli*. The phytochemical screening of the selected medicinal plants revealed diverse profiles of phytochemical components. Insights of these findings can stimulate further in-depth investigations to maximize the advantages that could be provided by these plant extracts for regular use as effective antimicrobial agents. The results confirm that the people of the study area have developed indigenous and local knowledge about the efficacies of the tested species through generations of repeated use. The research tested not only the plants but also the indigenous medical practice of the people of Habru District.

CHAPTER SIX

6. General Discussion, Conclusions and Recommendations

6.1. General discussion

This research aimed to examine the dynamics of land use and land cover (LULC) change, as well as the composition, structure, and analysis of plant communities within the Gerado, Micha, and Mekelet forest patches. Additionally, this study was intended to address the ethnobotanical study of traditional medicinal plants used in the Habru District, North Wollo Zone, Amhara Region, Ethiopia. The investigation also encompassed the evaluation of antibacterial activity and phytochemical screening of selected traditional medicinal plants most preferred by the indigenous and local people for treating infectious diseases.

The dynamics of LULC change and its driving forces are pivotal subjects in understanding the global environmental transformations induced by anthropogenic and natural factors (Belayneh Bufebo and Eyasu Elias, 2021). LULC changes are critical indicators of environmental shifts, reflecting the impacts of socio-economic development, urbanization, agricultural expansion, deforestation, and climate change on the earth's surface (Asnake Yimam and Amare Bantider, 2019). These changes significantly influence biodiversity, water and air quality, climate patterns, and carbon cycles, necessitating comprehensive studies to inform sustainable land management and policy decisions (Simachew Bantigegn, 2020). Studies conducted in Ethiopia have indicated that changes in Land Use and Land Cover (LULC) represent a critical challenge, exacerbating environmental issues (Asnake Yimam and Amare Bantider, 2019; Mehari Mariye *et al.*, 2022; Getahun Sisay *et al.*, 2023). In Ethiopia, LULC change mainly driven by various factors, including agricultural expansion, population growth, and biophysical factors has resulted in a declining vegetation cover and a serious deterioration of land quality over the past decades (EBI, 2022; Hailu Gisha et al., 2022; Getahun Sisay *et al.*, 2023). These changes are the results of a confluence of factors, encompassing a wide range of natural influences and socio-economic dynamics (Shiferaw Abate, 2011; Alemenesh Hailu *et al.*, 2020). Thus, monitoring and understanding these changes are essential for effective and sustainable land management and environmental conservation efforts in the country (Zhai *et al.*, 2020).

In the present study **(Chapter 2),** we assessed the land use land cover change dynamics and its driving factors in Habru District, North Wollo Zone of Amhara Region, Ethiopia. The study results revealed that five major land use and land cover types (water body, settlement area, bare land, cultivated land, shrub land, and forest area) were found in the study area. The study highlighted the dynamic changes in land use and land cover over the last 36 years, characterized by a marked increase in cultivated and settlement areas at the expense of natural vegetation (forest and shrub land). Such substantial expansions indicate a marked shift towards more intensive land use, particularly in terms of human settlement and agricultural activities, reflecting socio-economic development and possibly changes in water management practices that have led to the expansion of water bodies. The decrease in forest and shrub lands is particularly concerning, as these areas are crucial for biodiversity, carbon sequestration, and the provision of various ecosystem services (Alemenesh Hailu *et al.*, 2020). The results from the NDVI analysis also indicated a reduction in the vegetation cover within the study area, attributed to the deforestation for agricultural expansion and human settlement. Research results also reported that, the decreasing pattern of vegetation cover in Ethiopia is primarily driven by demand driven LULC change (EBI, 2022; Mesfin Anteneh, 2022), which is characterized by the expansion of agricultural land into marginal areas such as steep slopes, swampy plains, and traditionally untapped parts of the environment (Tsegaye Bekele, 2019). This expansion puts tremendous pressure on soil, vegetation, and water resources, leading to soil erosion, biodiversity loss, and land degradation (Mehari Mariye *et al.*, 2022). These findings underscore the dynamic nature of LULC changes and their implications for environmental sustainability and management. The increase in agricultural and urban areas at the expense of natural landscapes points to the need for integrated land use planning and conservation strategies. Such strategies should aim to balance developmental needs with environmental preservation, ensuring the sustainable use of land resources while mitigating negative impacts on ecosystems and biodiversity.

Moreover, the dynamic transformations in land use and land cover within the Habru District have critical implications for the diversity of woody species, as indicated by the substantial decline in forest and shrub lands. The reduction in these natural vegetative covers, primarily driven by agricultural expansion and settlement, results in a significant loss of biodiversity, adversely affecting the woody species composition. The changing LULC patterns, characterized by increased cultivation and human habitation, directly correlate with the observed shifts in woody species diversity. These alterations not only reduce the habitat available for various species but also disrupt the ecological balance necessary for sustaining diverse plant communities. It is essential to integrate sustainable land management practices that consider the conservation of these woody species to maintain ecological integrity and biodiversity (Alemenesh Hailu *et al.*, 2020; Getahun Sisay *et al.*, 2023).

In Chapter 3, we studied the woody species composition, structure, and plant community analysis of Gerado, Micha and Mekelet forest patches. The findings of the study identified 55 woody plant species belonging to 46 genera and 31 families and four plant community types significantly different ($R = 0.4703$, $P \leq 0.001$) in terms of floristic compositions, which are highly influenced by topographic variables (altitude, slope) and anthropogenic disturbance (tree cutting). Several research results also indicated that, the composition and distribution of species within plant communities are significantly influenced by topographic variables and anthropogenic disturbances (Crimmins *et al.*, 2008; Misganaw Meragiaw *et al.*, 2018; Rahman *et al.*, 2022; Gxasheka *et al.*, 2023; Metsehet Yinebeb *et al.*, 2023; Zeleke Assefa *et al.*, 2023). Topographic variables such as elevation and slope play a significant role in shaping plant community distribution patterns (Bisht *et al.*, 2022; Zeleke Assefa *et al.*, 2023). Moreover, human activities such as settlement, agricultural expansion, overgrazing by livestock, tree cutting, firewood collection, charcoal production and road construction can significantly affect the growth and distribution of plant communities, leading to changes in species richness, diversity, and composition (Liyew Birhanu *et al.*, 2021; Bentsi-Enchill *et al.*, 2022; Bisht *et al.*, 2022; Zeleke Assefa *et al.*, 2023). Our findings also revealed that. the differential impact of human activities across the forest patches, suggesting the necessity for sustainable conservation measures to address the specific challenges faced by each patch.

The result on the three forest patches (Gerado, Micha, and Mekelet) reveals a notable degree of similarity among them, with a mean similarity score of 0.65±0.08, attributed to their proximity and shared environmental influences (Friis *et al.*, 2011). Redundancy Analysis (RDA) highlighted significant relationships between community types within these patches and environmental variables, pinpointing altitude, slope, and tree cutting as pivotal in determining species composition and distribution. This aligns with findings from other Ethiopian studies emphasizing altitude's critical role in vegetation patterns (Haile Yineger *et al.*, 2011; Misganaw Meragiaw *et al.*, 2018; Admassu Addi *et al.*, 2020; Abera Anamo *et al.*, 2023; Metsehet Yinebeb *et al.*, 2023; Zeleke Assefa *et al.*, 2023). Species diversity and evenness across the four community type fell within accepted Shannon-Wiener index ranges, with Gerado exhibiting the highest species richness, followed by Micha and Mekelet, a variance likely influenced by both environmental and anthropogenic factors (Abyot Dibaba *et al.*, 2014; Misganaw Meragiaw *et al.*, 2018).

Furthermore, the study findings revealed that *Dodonaea viscosa* subsp. *angustifolia, Olea europaea* L. subsp. *cuspidata* and *Vachellia sieberiana* are the most dominant woody species found in the study forest patches. The dominance of *Dodonaea viscosa* subsp. *angustifolia* in the Gerado and Micha forest patches and *Vachellia sieberiana* in Mekelet, reflecting their adaptability to specific environmental conditions prevalent in dry Afromontane forests (DAF) of Ethiopian and semi-arid African landscapes (Harrington and Gadek, 2009; Hadgu Hishe *et al.*, 2021; Mubarek Eshetie *et al.*, 2021). Additionally, the presence of *Eucalyptus camaldulensis* in Mekelet points to varying degrees of human impact, distinguishing it from the other patches. These findings on species frequency and relative frequency (RF) underscore the ecological dominance of specific species due to their environmental resilience and the influence of anthropogenic factors, shaping forest structure and composition (Pohjonen and Pukkala, 1990; Shiferaw Alem and Pavlis, 2012). The study findings also demonstrate an inverted J-shape distribution in DBH and height classes, suggesting a stable population with a dominance of younger trees, as evidenced by (Birhanu Woldie *et al.*, 2015; Alemayehu Yig*Pittosporum viridiflorum*, *Dodonaea viscosa* subsp. *angustifolia*, and *Vachellia sieberiana* are the most ecologically significant species, highlighting their crucial role in the forest ecosystem's density, frequency, and canopy cover.

Also, the diversity of woody plant species in Gerado Micha and Mekelet forest patches plays a pivotal role in shaping the traditional and local knowledge concerning medicinal plants. The indigenous and local people of Habru District rely extensively on this biodiversity for their ethnomedicinal practices, as highlighted by the documentation of 35 out of 55 woody species also recognized for their medicinal use. The depth of ethnomedicinal knowledge is inherently tied to the richness of the local flora, where each species holds specific uses that have been passed down through generations. This study indicated that preserving woody plant species diversity is thus crucial not only for ecological reasons but also for maintaining and enhancing the repository of traditional knowledge. This traditional knowledge is integral for the sustainable use and potential discovery of new pharmacological agents, emphasizing the need for conservation strategies that protect these natural resources while fostering the continuity and enrichment of indigenous medicinal practices (Vivero *et al.*, 2005; Misganaw Meragiaw *et al.*, 2018).

The study also found that **(Chapter 4)** Habru District is rich in medicinal plant diversity as shown by the presence of 134 species belonging to 110 genera and 54 families for treating human ailments. Of these, two medicinal plant species (*M. ferruginea* and *U. simensis*) were recorded as endemic, depicting the area's distinct ecological and botanical attributes. These endemic plants are not only crucial for their potential therapeutic benefits but also enrich the field of ethnopharmacology, illustrating the significance of the local flora in traditional medicine practices (Vivero *et al.*, 2005; Hiwot Ayalew *et al.*, 2022).

The finding of this study also showed that the Solanaceae, Fabaceae, and Lamiaceae families are predominant in the Habru District, with the highest number of species identified as medicinal plants. The dominance of these species could be attributed to their wider distribution and abundance in the flora area (Haile Yineger *et al.*, 2007; Tilahun Teklehaymanot, 2009; Mirutse Giday *et al.*, 2010; Ermias Lulekal *et al.*, 2013; Gadisa Demie *et al.*, 2018). The number of reported medicinal plants in the district found to be higher than that of other adjacent districts investigated to document the ethnomedicinal wealth (Tilahun Teklehaymanot, 2017; Tibebu Tefera and Mesele Yihune, 2019) and lower than another study by Getnet Chekole (2017). These studies collectively illustrate a significant botanical heritage in the Habru District and its adjacent areas, with specific plant families like Fabaceae and Asteraceae frequently documented across different regions (Haile Yineger *et al.*, 2007; Alemayehu Kefalew *et al.*, 2015; Gadisa Demie *et al.*, 2018; Netsanet Gonfa *et al.*, 2020; Mikiyas Abebe, 2022). Furthermore, the consistent findings across the different regions highlight the importance of these plant families in traditional medicinal systems, contributing to a broader understanding of the area's ethnobotanical knowledge. This similarity in the diversity of medicinal plant in adjacent areas of a country could be attributed to the shared ecological and environmental conditions as well as ethnomedicinal knowledge and practices among local communities within these geographically adjacent districts (Friis *et al.*, 2011; Ermias Lulekal *et al.*, 2013). Additionally, the extensive use of species from these families could be linked to the presence of effective bioactive compounds against diseases, as previously described in other studies (Gazzaneo *et al.*, 2005; Elizalde-Romero *et al.*, 2021; Obistioiu *et al.*, 2021; Alagar Yadav and Sara Koshi, 2022; Grygier *et al.*, 2022; da Silva *et al.*, 2023).

The study highlights that in the Habru District, herbs constitute the majority of medicinal plants (36%), followed by shrubs (32%) and trees (25%), suggesting that herbs' predominance may be due to their easier accessibility compared to trees and shrubs, which are typically found in more distant forested areas. This pattern of herbaceous species being more commonly used as medicinal plants aligns with other studies in Ethiopia and elsewhere (Tabuti *et al.*, 2003; Haile Yineger *et al.*, 2007; Mirutse Giday *et al.*, 2010; Ermias Lulekal *et al.*, 2013). The study reveals that despite the district's wild habitats hosting a diversity of medicinal plants, human activities, including tree cutting, collecting firewood, and constructing houses, have resulted in the loss of important medicinal plant species, a similar result also documented by Haile Yineger *et al.* (2007); Ermias Lulekal *et al.* (2013). According to the local people’s knowledge on remedy preparation, leaves are the most widely used plant part followed by roots and other plant parts, indicating the depth of traditional medicinal practices, aligning with studies across Ethiopia that highlight leaves as the most used plant part due to their high concentration of bioactive compounds (Ketema Tolossa *et al.*, 2013; Balcha Abera, 2014; Hiwot Ayalew *et al.*, 2022). The significant reliance on root harvesting for medicinal plant preparations, presents a sustainability challenge for the survival of these plants, with similar concerns reported across various ethnomedicinal studies in Ethiopia (Haile Yineger *et al.*, 2007; Tilahun Teklehaymanot, 2009; Ermias Lulekal *et al.*, 2013; Balcha Abera, 2014; Zewdie Kassa *et al.*, 2020). This extraction method, particularly when involving the complete removal of roots, threatens the plants' ability to survive, especially those that are rare and reproduce slowly. In this study, leaf harvesting for remedies, also a common practice, offers a more sustainable alternative, allowing plants to continue their growth and functions. Thus, promoting leaf collection over root harvesting could significantly contribute to the conservation of medicinal plant species.

This research also indicates that fresh plant parts, comprising 58.2% of the materials, were predominantly utilized in preparing remedies for a variety of health conditions. This preference for fresh over dried plant materials is attributed to the belief in their superior therapeutic effectiveness, owing to higher concentrations of active compounds that might diminish upon drying. This result aligns with similar research findings emphasizing the enhanced efficacy of fresh plant parts in traditional medicine (Mirutse Giday *et al.*, 2010; Zewdie Kassa *et al.*, 2020; Mersha Ashagre and Ermias Lulekal, 2021). Oral application emerged as the primary route of administration, reflecting a broader trend within Ethiopian traditional medicine (Ermias Lulekal *et al.*, 2013; Getnet Chekole, 2017; Zewdie Kassa *et al.*, 2020). The market survey result indicated that among the reported medicinal plants only 22 (16.4%) medicinal plants species are marketable, with only a few species (*T. brownii*, *M. communis*, *R. chalepensis*, *O. europaea* L. subsp. *cuspidata*, *A. sativum* and *C. annum)* found in local markets. The market value of these species highlights their potential for income generation and community demand but also suggests they may face pressure due to targeted harvesting for economic gain (Ermias Lulekal *et al.*, 2013; Mersha Ashagre and Ermias Lulekal, 2021).

The study found high Informant Consensus Factor (ICF) values of 0.85 and 0.84 for medicinal plants used in treating gastrointestinal, parasitic ailments, and febrile diseases, indicating strong agreement among informants on their use. This consensus not only highlights the cultural and practical relevance of these plants within traditional healing practices but also emphasizes the need for their conservation and further research into their pharmacological properties (Heinrich *et al.*, 1998). Consequently, 23 medicinal plants from the Habru District, identified for their notable ICF values, are currently being researched for their pharmacological efficacy against gastrointestinal and parasitic diseases.

The finding in this study on the highest fidelity level (FL) values for *S. somalense* (91.3%) in treating gastro-intestinal and parasitic diseases and *O. lamiifolium* (88.9%) for febrile diseases; and *V. sinaiticum* (85.7%) for wound, highlights the relative therapeutic potential of these medicinal plants. These plants with high FL value could be considered as prime candidates for further phytochemical research to identify active compounds and for conservation efforts due to their significant medicinal or cultural value (Tangjitman *et al.*, 2015). The preference ranking exercise also pinpointed the top medicinal plant species for treating diarrheal diseases within the gastrointestinal and parasitic disease category. Accordingly, *S. somalense* (YESHEHOCHU KITEL), *C. aurea* (DIGITA.) and *V. sinaiticum* (YEAHIYA JORO) emerged as the most preferred species to treat diarrheal diseases in the study area. The results of DMR exercise identified *O. europaea* subsp. *cuspidata*, *D. viscosa* subsp. *angustifolia*, and *E. racemosa* as the top-ranked multipurpose plant species. The results of DMR exercise reveal that the primary exploitation of *O. europaea* subsp. *cuspidata*, *D. viscosa* subsp. *angustifolia* and *E. racemosa* leans more towards their non-medicinal applications, such as house construction and firewood, rather than their medicinal properties. This tendency towards overharvesting for non-medicinal uses is pinpointed as a key factor contributing to the species' depletion in the study area, as noted earlier (Haile Yineger *et al.*, 2007; Yimer Assen *et al.*, 2021; Moa Megersa and Samuel Woldetsadik, 2022). Consequently, the research underscores the urgent necessity for implementing complementary conservation strategies to preserve these rapidly diminishing multipurpose medicinal plant species in the study area (Haile Yineger *et al.*, 2007; Yimer Assen *et al.*, 2021; Moa Megersa and Samuel Woldetsadik, 2022). Furthermore, the study result showed that 11 medicinal plants were identified as a wild edible plant in the district. The interconnection between medicinal and edible plants offers insights into their nutraceutical role, contributing to food security and showcasing the rich traditional knowledge within local communities, a theme explored in various cultures and regions (Aparicio *et al.*, 2021). Thus, further research is needed to explore the nutritional and medicinal properties of specific plant species and their potential for sustainable food systems (Haile Tesfaye, 2020; Muhidin Tahir *et al.*, 2021; Mulatu Hankiso *et al.*, 2023).

The study indicates the local community's opportunities, such as enhancing awareness, strengthening research and development, and supporting traditional healers, paving the way for better use and regulation of herbal medicines. Conversely, it also points out significant challenges, including deforestation, agricultural expansion, and environmental degradation, which threaten the sustainability of medicinal plant resources and call for dedicated conservation strategies. The research reveals a complex relationship between biodiversity, cultural practices, and healthcare systems in Habru District, offering a rich base for further scientific inquiry, preservation efforts, and possible integration into contemporary healthcare system. Comparative analysis with other ethnobotanical studies in Ethiopia uncovered unique applications of certain medicinal plants in the district. Notably, the use of *S. somalense* for a variety of conditions such as headache, fever, and diarrhea, and the novel application of *C. gynandra* for wounds and ear infections, previously unreported, highlight the distinctive medicinal plant usage in Habru. The documented species and their traditional knowledge hold significant promise for future public health initiatives and the evolution of sustainable herbal medicine practices in the district.

Accordingly, the conservation of medicinal woody species in forest patches like Gerado, Micha, and Mekelet holds significant pharmacological advantages. Many of these species contain bioactive compounds that have been traditionally used to treat various ailments, and their conservation ensures the continued availability of these natural resources for pharmaceutical research and drug development. The study's findings on the antibacterial properties and phytochemical screening of plants underline the medical relevance of these species. By protecting these habitats and the species they harbor, there is an opportunity to harness their potential for developing new treatments, thereby contributing to the global medicinal reservoir, and promoting sustainable health solutions (Ermias Lulekal *et al.*, 2013; Shemsu Umer *et al.*, 2013).

Chapter 5 delves into the study of antibacterial effects and the phytochemical composition of favored traditional medicinal plants (*S. somalense, V. sinaiticum, R. nervosus, W. somnifera*, and *C. aurea*) that are most preferred for treating infectious diseases. The study found that ethanolic extracts from *S. somalense, W. somnifera*, and *C. aurea*, plants with high informant consensus and fidelity levels, have antibacterial properties against various bacterial strains. *S. somalense* extract was effective against *S. agalactiae, K. pneumoniae*, and *E. coli*. Whereas *W. somnifera* extract showed antibacterial activity against *S. aureus* and *E. coli*. Multiple investigations into antibacterial effectiveness and phytochemical assessments have consistently shown positive outcomes that support traditional medicinal practices (Hailu Tadeg *et al.*, 2005; Shemsu Umer *et al.*, 2013; Dharajiya *et al.*, 2014; Ermias Lulekal *et al.*, 2014; Hazman *et al.*, 2021; Kalkidan Lulseged *et al.*, 2022; Minda Asefa *et al.*, 2022). Similar study reported that, methanol extracts of *W. somnifera* (Dharajiya *et al.*, 2014), methanol extracts of *R. nervosus* (Al Yahya et al., 2018), methanol extract of *C. aurea* (Gebremedhin Romha *et al.*, 2018; Dessie Belay *et al.*, 2021; Solomon Berhanu, 2022) were effective against both against various bacterial strains, contributing to the growing evidence of the plant's antibacterial properties. The phytochemical analysis of ethanolic extracts from five medicinal plants revealed a broad spectrum of secondary metabolites, with all extracts testing positive for steroids. Specifically, *S. somalense* was rich in steroids, alkaloids, flavonoids, saponins, and terpenoids, showcasing its diverse bioactive compound profile. The study also revealed that the plant extracts contain a diverse array of phytochemicals, such as phenolic compounds, saponins, flavonoids, and alkaloids. These substances are key contributors to the antibacterial capabilities of *C. aurea*. Earlier reports also confirmed that an 80% methanol extract from the leaves of *C. aurea* demonstrated antimicrobial effects against several pathogens, including *Salmonella typhimurium*, *various Shigella* species, *P. aeruginosa, S. aureus*, and *E. coli* (Shemsu Umer *et al.*, 2013). The observed antibacterial efficacy of tested extracts, alongside the identified bioactive compounds, underscores their potential for drug development, supporting their traditional medicinal use. Specifically, the alignment of *S. somalense*'s bioactive components with its antibacterial capabilities, and the chemical diversity in *W. somnifera* and *C. aurea*, validates their historical use in treating infections.

6.2. Conclusions

This study presented a quantitative data analysis of woody plant species composition, structure, and plant community analysis in relation to land use land cover (LULC) change dynamics, and ethnobotanical study of medicinal plants including antimicrobial activity testing of most preferred healing plants in Habru District. The analysis of LULC change dynamics within Habru District reveals significant shifts over the past few decades (1985 - 2021), driven primarily by socio-economic developments and population growth. The increase in cultivated lands, settlement areas, and water bodies at the expense of shrub and forest lands underscores the impact of human activities and climatic variations on the environment. This LULC transformation highlights the intricate balance between human livelihood needs and ecosystem conservation, necessitating the implementation of integrated land management strategies to mitigate adverse impacts on biodiversity and ecosystem services.

The findings of this study revealed that, a total of 55 woody plant species belonging to 46 genera and 31 families from the three forest patches distributed in four plant community types were recorded. The study identifies the profound impacts of anthropogenic disturbances including activities such as tree cutting, firewood collection, and charcoal production on species richness, vegetation structure and plant community characteristics. By linking the findings from these forest patches with the broader LULC changes, it becomes evident that sustainable management and conservation strategies must consider both the ecological and socio-economic dimensions to foster resilience against environmental changes and anthropogenic pressures.

This study also reveals a deep connection between the local community and medicinal plant species, highlighting the critical role of traditional knowledge in healthcare. A total of 134 medicinal plant species in 110 genera and 54 families, including 2 endemics used for 50 human ailments were recorded. This result underscores the region's biodiversity and its significance for conservation and medicinal potential.

Documenting medicinal plants by their use value, preference ranking scores, and fidelity level values could enhance the effectiveness of future pharmaceutical and phytochemical research, as well as support conservation efforts. This study investigated the antibacterial activity and phytochemical screening of selected medicinal plants, confirming their efficacy against certain gram positive and gram-negative bacterial strains. This finding validates the traditional use of these plants in treating infectious diseases and opens avenues for pharmaceutical research. Accordingly, the study found that extracts from *Solanum somalense* Franchet., *Withania somnifera* (L.) Dunal and *Calpurnia aurea* (Ait.) Benth. demonstrated antimicrobial activity against pathogens like *Staphylococcus aureus, Streptococcus agalactiae, Klebsiella pneumoniae* and *Escherichia coli*, with MICs ranging from 1.5 to 3.5 mg/mL, supported by phytochemical analyses revealing diverse bioactive compounds. These results, validating the indigenous medical knowledge of Habru District's residents, suggest the potential of these plant extracts as effective antimicrobial agents and warrant further detailed research. Moreover, this finding emphasizes the value of integrating traditional knowledge with scientific research to harness the therapeutic potentials of biodiversity, thereby contributing to both public health and biodiversity conservation objectives.

This study underlines the intricate interplay between land cover dynamics, plant diversity, and human interaction in Habru District. It highlights the urgency of adopting sustainable land management, biodiversity conservation, and the integration of traditional knowledge into modern primary healthcare practices and education. These findings contribute significantly to the understanding of ecological and ethnobotanical dynamics in Habru District, offering a basis for future conservation and sustainable use of these vital resources.

6.3. Recommendations

Based on the research results, the following recommendations are proposed:

- ✓ **Sustainable land use and conservation practices**: Given the significant land cover changes observed, it is crucial to implement sustainable land management and biodiversity conservation strategies. Efforts should focus on controlling the expansion of settlement areas and cultivated land to prevent further depletion of shrub and forest lands. Improving the existed community-based forest patch conservation initiatives are essential to maintain the ecological balance and preserve natural resources. To reduce forest patch conversion, relevant government bodies like local governments at district level, the Ethiopian Forestry Development, Ethiopian Biodiversity Institute and the Ethiopian Ministry of Agriculture and Natural Resources should work together to introduce alternative means of income generation for local communities.
- ✓ **Policy implementation and environmental governance:** The local government at district level should apply the land tenure security, regulation of resource utilization, and the implementation of legal frameworks to protect biodiversity and traditional knowledge systems as well as mitigating the ramifications of LULC changes on the study area's vegetation cover and environment.
- ✓ **Preservation of woody species and plant communities**: The study of woody species composition and structure in Gerado Micha and Mekelet forest patches underscores the need for targeted conservation of these biodiversity-rich areas. The regional government of the study area and the district itself should take the initiatives including monitoring and preserving distinct plant communities, managing human disturbances, and promoting reforestation and afforestation programs to restore degraded lands.
- ✓ **Documentation and conservation of ethnobotanical knowledge**: The rich ethnobotanical knowledge documented in Habru District highlights the importance of preserving this cultural heritage. Armauer Hansen Research Institute (AHRI), Traditional and Modern Medicine Research and Development Directorate under Ministry of Health should design and implement strategies that include the documentation and conservation of traditional

medicinal practices, promoting the sustainable use of medicinal plants, and integrating this knowledge into modern healthcare systems.

- ✓ **Research and development of medicinal plants**: The study on antibacterial activity and phytochemical screening of medicinal plants suggests the potential for pharmaceutical development. Research institutions such as AHRI should prioritize *Solanum somalense* Franchet for further phytochemical investigation based on their high informant consensus and fidelity level values.

- ✓ **Community engagement and education**: It is vital to engage local communities in conservation efforts. The Ethiopian Biodiversity Institute (EBI) and Habru district environmental protection and forest resource conservation department in cooperation with the Ministry of Health should organize educational programs aimed at raising awareness about the importance of biodiversity conservation, sustainable resource use, and traditional knowledge on ethnobotanical uses of medicinal plants can foster a sense of stewardship and participation in conservation activities.

- ✓ **Prioritize conservation efforts፡** The responsible bodies, including the local government at district level, Ethiopian Biodiversity Institute, the Ethiopian Forestry Development, Ministry of Health, and the local communities should prioritize conservation efforts for highly marketable medicinal plants (*Terminalia brownii* Fresen. and *Myrtus communis*) and multipurpose plants (*O europaea* L. subsp. *cuspidata D. viscosa* subsp. *angustifolia* and *E. racemosa*) that are facing threats beyond their medicinal value.

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Appendices

Appendix 1: List of woody and medicinal plant species collected in Habru District, including GPS coordinates and voucher numbers

Scientific names	Local name (Amharic)	Family name	GF	Altitude	Latitude (dd mm ss)	Longitude (dd mm ss)	VS number	Remark
Acacia decurrens (J.C.Wendl.) Willd.	Akacha	Fabaceae	T	1956	11°44'25"	39°37'46"	MA135	2
Achyranthes aspera L.	Telenj	Amaranthaceae	H	1875	11°32'31"	39°37'03"	MA05	1
Afrocarpus falcatus (Thunb.) C.N.Page	Zigba	Podocarpaceae	T	2138	11°44'09"	39°37'49"	MA91	1,2
Agave sisalana Perrine	Chiret, Kacha	Asparagaceae	S	1872	11°32'27"	39°37'03"	MA06	1
Ajuga integrifolia Buch.-Ham. Ex D. Don	Armagusa/Dem Akurt	Lamiaceae	H	1871	11°32'19"	39°36'59"	MA129	1
Albizia gummifera (J.F.Gmel.) C.A.Sm.	Sesa	Fabaceae	T	1956	11°44'25"	39°37'46"	MA136	2
Allium sativum L.	Nech Shnkurt	Amaryllidaceae	H	1975	11°32'14"	39°36'36"	MA08	1
Allophylus abyssinicus (Hochst.) Radlk.	Embis	Sapindaceae	T	2198	11°46'42"	39°33'44"	MA137	2
Aloe macrocarpa Tod.	Iret / Iret tafa	Asphodelaceae	H	1871	11°32'19"	39°36'59"	MA09	1
Artemisia absinthium L.	Arity, Natra	Asteraceae	H	1563	11°34'03"	39°40'24"	MA10	1
Artemisia abyssinica Sch.Bip. ex Oliv. & Hiern	Chikugn	Asteraceae	H	1975	11°32'14"	39°36'36"	MA11	1
Arundo donax L.	Shenbeko	Poaceae	S	2108	11°44'15"	39°37'30"	MA138	2
Asparagus africanus Lam.	Yeset Kest, Kestanicha	Asparagaceae	H	1872	11°32'34"	39°37'04"	MA133	1
Balanites aegyptiaca (L.) Delile	Bedena	Zygophyllaceae	T	1644	11°35'42"	39°39'33"	MA12	1
Barleria eranthemoides R.Br.ex C.B.Clarke	Yeset Af	Acanthaceae	H	1873	11°32'29"	39°37'02"	MA13	1
Bidens pilosa L.	Yeseytan Merfe	Asteraceae	H	1873	11°32'29"	39°37'02"	MA14	1
Calotropis procera (Aiton) W.T.Aiton	Tobia	Apocynaceae	S	1945	11°44'24"	39°38'17"	MA15	1,2
Calpurnia aurea (Ait.) Benth.	Digita	Fabaceae	S	1930	11°44'23"	39°38'01"	MA16	1,2
Capparis decidua (Forssk.) Edgew.	Kontir	Capparaceae	S	1698	11°41'03"	39°38'18"	MA17	1
Capparis tomentosa Lam.	Gimero	Capparaceae	S	1966	11°32'18"	39°36'05"	MA18	1
Capsicum annuum L.	Karia	Solanaceae	H	1975	11°32'14"	39°36'36"	MA19	1
Capsicum frutescens L.	Mitmita Karia	Solanaceae	H	1975	11°32'14"	39°36'36"	MA20	1
Carica papaya L.	Papaya	Caricaceae	T	1975	11°32'14"	39°36'36"	MA21	1
Carissa spinarum L.	Agam	Apocynaceae	S	2014	11°44'21"	39°38'27"	MA22	1,2
Casuarina equisetifolia L.	Shiwshiwa	Casuarinaceae	T	1956	11°44'25"	39°37'46"	MA139	2

Catha edulis (Vahl) Endl.	Chat	Celastraceae	H	1867	11°32'23"	39°36'58"	MA23	1
Celtis africana Burm.f.	Awrarise/Ameleka	Cannabaceae	T	1956	11°44'25"	39°37'46"	MA25	1,2
Cicer arietinum L.	Shimbira	Fabaceae	H	1867	11°32'23"	39°36'58"	MA26	1
Cissus quadrangularis L.	Yezhon Anjet	Vitaceae	C	1872	11°32'34"	39°37'04"	MA27	1
Citrus aurantiifolia (Christm.) Swingle	Lomi	Rutaceae	T	1970	11°44'15"	39°38'12"	MA28	1,2
Citrus medica L.	Tiringo	Rutaceae	T	1703	11°40'52"	39°38'58"	MA29	1
Clematis simensis Fresen.	Nech Yeazo-hareg	Ranunculaceae	C	2004	11°32'16"	39°36'03"	MA30	1
Cleome gynandra L.	Abethoye	Cleomaceae	H	1644	11°35'42"	39°39'33"	MA31	1
Coffea arabica L.	Bunna	Rubiaceae	S	1867	11°32'23"	39°36'58"	MA33	1
Cordia africana Lam.	Wanza	Boraginaceae	T	2108	11°44'15"	39°37'30"	MA34	1,2
Croton macrostachyus Hochst. ex Delile	Bisana	Euphorbiaceae	T	2099	11°46'41"	39°33'34"	MA35	1,2
Cucumis ficifolius A.Rich.	Yemidir Embway	Cucurbitaceae	H	1966	11°32'17"	39°36'05"	MA36	1
Cucurbita pepo L.	Duba	Cucurbitaceae	C	1644	11°35'42"	39°39'33"	MA37	1
Cymbopogon martini (Roxb.) Will.Watson	Teje Sar	Poaceae	H	1875	11°44'46"	39°37'09"	MA38	1
Cynoglossum coeruleum Hochst. ex A.DC.	Fikru-tena, Chigogot	Boraginaceae	H	2054	11°32'24"	39°36'15"	MA132	1
Cyphostemma adenocaule (Steud. ex A.Rich.) Desc. ex Wild & R.B.Drumm.	Aserkush Tebetebkush	Vitaceae	C	1872	11°32'34"	39°37'04"	MA24	1
Datura stramonium L.	Astenagir/Atsefaris	Solanaceae	S	1877	11°32'39"	39°37'04"	MA39	1
Dichrostachys cinerea (L.) Wight & Arn.	Awrarise/Ader/Gorgoro	Fabaceae	S	2158	11°47'04"	39°31'52"	MA40	1,2
Dodonaea viscosa subsp. *angustifolia* (L.f.) J.G.West	Kitkita	Sapindaceae	S	2069	11°46'48"	39°33'36"	MA41	1,2
Dombeya torrida (J.F.Gmel.) Bamps	Danisa/Kuncho	Malvaceae	S	2008	11°47'21"	39°32'03"	MA140	2
Ehretia cymosa Thonn.	Wulaga	Boraginaceae	T	1956	11°44'25"	39°37'46"	MA42	1,2
Embelia schimperi Vatke	Enkoko	Primulaceae	S	2054	11°32'24"	39°36'15"	MA43	1
Eucalyptus camaldulensis Dehnh.	Key Bahirzaf	Myrtaceae	T	2083	11°47'15"	39°32'02"	MA44	1,2
Eucalyptus globulus Labill.	Nech Bahir Zaf	Myrtaceae	T	2589	11°43'27"	39°33'41"	MA45	1
Euclea racemosa L.	Dedeho	Ebenaceae	S	2214	11°46'30"	39°33'24"	MA46	1,2
Euphorbia abyssinica J.F.Gmel.	Kulkual	Euphorbiaceae	S	1930	11°44'28"	39°38'01"	MA47	1,2
Euphorbia tirucalli L.	Kinchibt	Euphorbiaceae	S	2017	11°47'26"	39°32'27"	MA48	1,2
Faidherbia albida (Delile) A.Chev.	Gerbi	Fabaceae	T	2169	11°44'13"	39°37'36"	MA141	2
Ferula communis L.	Dog	Apiaceae	H	1867	11°32'23"	39°36'58"	MA49	1
Ficus capreifolia Delile	Beles	Moraceae	T	2012	11°47'26"	39°32'19"	MA142	2

Ficus carica L.	Beles	Moraceae	T	2054	11°32'24"	39°36'15"	MA50	1
Ficus sur Forssk.	Sholla	Moraceae	T	2008	11°47'21"	39°32'03"	MA51	1,2
Ficus vasta Forssk.	Warka	Moraceae	T	2056	11°44'18"	39°37'34"	MA52	1,2
Foeniculum vulgare Mill	Ensilal	Apiaceae	H	1867	11°32'23"	39°36'58"	MA53	1
Galiniera saxifraga (Hochst.) Bridson	Buna Mesay	Rubiaceae	S	1991	11°44'20"	39°38'17"	MA143	2
Grevillea robusta A.Cunn. ex R.Br.	Gravilea	Proteaceae	T	2108	11°44'15"	39°37'30"	MA144	2
Grewia bicolor Juss.	Sefa	Malvaceae	S	1929	11°44'27"	39°37'55"	MA54	1,2
Grewia ferruginea Hochst. ex A.Rich.	Lenkuata	Malvaceae	S	2081	11°46'46"	39°33'15"	MA55	1,2
Grewia villosa Willd.	Agobday	Malvaceae	S	2053	11°32'24"	39°36'11"	MA56	1
Guizotia abyssinica (L.f.) Cass.	Nug	Asteraceae	H	1644	11°35'42"	39°39'33"	MA57	1
Gymnanthemum amygdalinum (Delile) Sch.Bip.	Girawa	Asteraceae	T	2046	11°47'09"	39°33'15"	MA122	1,2
Gymnosporia senegalensis (Lam.) Loes.	Atat	Celastraceae	S	2097	11°47'10"	39°31'59"	MA73	1,2
Hesperocyparis lusitanica (Mill.) Bartel	Yeferenj Tsid	Cupressaceae	T	2046	11°47'09"	39°31'46"	MA145	2
Heteromorpha arborescens (Spreng.) Cham. & Schltdl.	Yejib Mirkuz	Apiaceae	S	2021	11°44'16"	39°37'56"	MA146	2
Hordeum vulgare L.	Gebs	Poaceae	H	1644	11°35'42"	39°39'33"	MA58	1
Jacaranda mimosifolia D.Don	Yetebenja Zaf /Jakaranda	Bignoniaceae	T	1644	11°35'42"	39°39'33"	MA147	2
Jasminum abyssinicum Hochets. Ex DC.	Tenbelel	Oleaceae	C	1874	11°32'20"	39°39'36"	MA60	1
Juniperus procera Hochst. ex Endl.	Yehabesha Tsid	Cupressaceae	T	2093	11°47'07"	39°31'49"	MA61	1,2
Justicia schimperiana (Hochst. ex Nees) T.Anderson	Sensel, Simiza	Acanthaceae	S	1854	11°32'39"	39°37'10"	MA62	1
Kalanchoe petitiana A.Rich.	Endahula	Crassulaceae	H	1854	11°32'39"	39°37'10"	MA63	1
Lagenaria siceraria (Molina) Standley	Qil	Cucurbitaceae	C	1739	11°43'05"	39°39'01"	MA64	1
Lantana camara L.	Yewef Kolo	Verbenaceae	S	1853	11°32'34"	39°37'10"	MA65	1
Lantana trifolia L.	Yeregna Kolo	Verbenaceae	S	1949	11°32'17"	39°36'03"	MA66	1
Leonotis ocymifolia (Burml. f.) Iwarsson	Geram Tinjut, Eraskimir	Lamiaceae	S	1853	11°32'34"	39°37'10"	MA130	1
Lepidium sativum L.	Feto	Brassicaceae	H	1853	11°32'34"	39°37'10"	MA67	1
Linum usitatissimum L.	Telba	Linaceae	H	1853	11°32'34"	39°37'10"	MA68	1
Lippia abyssinica (Otto & A.Dietr.) Cufod.	Kessie	Verbenaceae	S	1851	11°32'38"	39°37'09"	MA69	1
Loranthella deflersii (Tiegh.) S.Blanco & C.E.Wetzel	Yebedena Tegedra	Loranthaceae	H	1851	11°32'39"	39°37'10"	MA85	1

Malva verticillata L.	Lut/Adguar	Malvaceae	C	2044	11°32'23"	39°36'21"	MA71	1
Mangifera indica L.	Mango	Anacardiaceae	T	1623	11°33'28"	39°39'25"	MA72	1
Millettia ferruginea (Hochst.) Hochst. ex Baker	Birbira	Fabaceae	T	1874	11°44'47"	39°37'10"	MA74	1
Momordica foetida Schumach.	Yekura Hareg	Cucurbitaceae	C	1949	11°32'17"	39°36'03"	MA75	1
Moringa oleifera Lam.	Shiferaw/Moringa	Moringaceae	T	1623	11°33'28"	39°39'25"	MA76	1,2
Myrsine africana L.	Kechemo	Primulaceae	S	2226	11°47'00"	39°31'56"	MA77	1,2
Myrtus communis L.	Ades	Myrtaceae	S	1563	11°34'03"	39°40'24"	MA78	1
Nicotiana tabacum L.	Tinbaho	Solanaceae	S	1975	11°32'14"	39°36'36"	MA79	1
Nigella sativa L.	Tikur Azmud	Ranunculaceae	H	1644	11°35'42"	39°39'33"	MA80	1
Ocimum basilicum L.	Besobila	Lamiaceae	H	1783	11°43'06"	39°39'01"	MA81	1
Ocimum gratissimum (L.) subsp. *gratissimum*	Damakasse, Kesedama	Lamiaceae	S	1874	11°32'19"	39°36'58"	MA83	1
Ocimum lamiifolium Hochst. ex. Benth.	Dama Kessie	Lamiaceae	H	1851	11°32'39"	39°37'10"	MA82	1
Olea europaea L. subsp. *cuspidata* (Wall. & G.Don) Cif.	Weira	Oleaceae	T	2138	11°44'09"	39°37'49"	MA84	1,2
Olinia rochetiana A. Juss.	Tfie	Penaeaceae	S	1876	11°32'19"	39°36'58"	MA131	1
Opuntia ficus-indica (L.) Mill.	Qulqual / Manka/Beles/Wedi	Cactaceae	S	2169	11°44'13"	39°37'36"	MA86	1,2
Osyris lanceolata Hochst. & Steud.	Keret	Santalaceae	S	2169	11°44'13"	39°37'36"	MA87	1,2
Periploca linearifolia Quart.-Dill. & A.Rich.	Moider	Apocynaceae	C	1876	11°32'19"	39°36'58"	MA111	1
Persea americana Mill.	Avocado	Lauraceae	T	1860	11°32'37"	39°37'10"	MA88	1
Physalis lagascae Roem. & Schult.	Awut	Solanaceae	H	1851	11°32'39"	39°37'10"	MA89	1
Phytolacca dodecandra L'Hér.	Endod	Phytolaccaceae	S	2004	11°32'16"	39°36'02"	MA90	1
Pittosporum viridiflorum Sims	Kefeto	Pittosporaceae	T	2087	11°47'09"	39°31'52"	MA148	2
Premna schimperi Engl.	Checho	Lamiaceae	S	2413	11°46'21"	39°33'25"	MA149	2
Prunus africana (Hook.f.) Kalkman	Tiqur Inchet	Rosaceae	T	2087	11°47'09"	39°31'52"	MA92	1
Psidium guajava L.	Zeyetun	Myrtaceae	T	1856	11°44'47"	39°37'09"	MA93	1
Pterolobium stellatum (Forssk.) Brenan	Kentafa	Fabaceae	S	2014	11°47'15"	39°31'52"	MA94	1,2
Rhamnus prinoides L'Hér.	Gesho	Rhamnaceae	S	1991	11°44'19"	39°38'07"	MA95	1,2
Ricinus communis L.	Gulo	Euphorbiaceae	S	1876	11°32'19"	39°36'58"	MA98	1
Rosa abyssinica R.Br. ex Lindl.	Kega	Rosaceae	S	2100	11°47'15"	39°32'09"	MA99	1,2
Rotheca myricoides (Hochst.) Steane & Mabb.	Misiroch	Lamiaceae	H	1931	11°32'23"	39°36'19"	MA32	1
Rubia cordifolia L.	Mencherer	Rubiaceae	H	1944	11°32'23"	39°36'09"	MA100	1

Rubus fruticosus L.	Enjori	Rosaceae	S	1872	11°32'34"	39°37'04"	MA101	1
Rumex abyssinicus Jacq.	Mekmeko	Polygonaceae	H	1876	11°32'19"	39°36'58"	MA102	1
Rumex nepalensis Spreng.	Kitel Rejim; Tult	Polygonaceae	H	1872	11°44'48"	39°37'11"	MA103	1
Rumex nervosus Vahl	Embacho	Polygonaceae	S	1872	11°44'48"	39°37'11"	MA104	1
Ruta chalepensis L.	Tenadam	Rutaceae	H	1876	11°32'19"	39°36'58"	MA105	1
Saccharum officinarum L.	Shenkora Ageda	Poaceae	T	1867	11°32'23"	39°36'58"	MA106	1
Salvia nilotica Juss. ex Jacq.	Hulgeb	Lamiaceae	H	1872	11°32'34"	39°37'04"	MA07	1
Schinus molle L.	Kundo-berberie	Anacardiaceae	T	2012	11°47'26"	39°32'19"	MA107	1,2
Searsia glutinosa (Hochst. ex A.Rich.) Moffett	Embis	Anacardiaceae	S	1977	11°44'18"	39°38'27"	MA150	2
Searsia pyroides (Burch.) Moffett	Yeahya Talo, Yeregna Qolo	Anacardiaceae	S	1625	11°33'26"	39°39'26"	MA97	1
Searsia retinorrhoea (Steud. ex Oliv.) Moffett	Talo/Talo Embis	Anacardiaceae	T	2198	11°46'34"	39°33'18"	MA96	1,2
Senegalia brevispica (Harms) Seigler & Ebinger	Kontr	Fabaceae	S	2037	11°47'13"	39°31'55"	MA151	2
Senegalia senegal (L.) Britton	Sibansa Grar	Fabaceae	T	1644	11°35'42"	39°39'33"	MA02	1
Sida schimperiana Hochst. ex A. Rich.	Chifrig	Malvaceae	S	1982	11°32'16"	39°36'03"	MA108	1
Sisymbrium officinale (L.) Scop.	Yewef Gomen	Brassicaceae	H	1644	11°35'42"	39°39'33"	MA109	1
Solanum anguivi Lam.	Zirch-embuay	Solanaceae	H	1975	11°32'15"	39°36'03"	MA110	1
Solanum dasyphyllum Schumach. & Thonn.	Geber-embuay	Solanaceae	S	1875	11°32'31"	39°37'03"	MA135	1
Solanum incanum L.	Embuay	Solanaceae	S	1872	11°32'34"	39°37'04"	MA112	1
Solanum lycopersicum L.	Timatim	Solanaceae	H	1783	11°43'06"	39°39'01"	MA70	1
Solanum marginatum L.f.	Embuay	Solanaceae	S	1872	11°32'34"	39°37'04"	MA113	1
Solanum somalense Franchet.	Yeshehochu Kitel /Shejerete Jin	Solanaceae	S	1619	11°35'42"	39°39'32"	MA114	1
Stephania abyssinica (Quart.-Dill. & A.Rich.) Walp.	Yeayit Joro/Etse Eyesus	Menispermaceae	C	1875	11°44'46"	39°37'09"	MA115	1
Tapinanthus globifer (A.Rich.) Tiegh.	Yebuna Tegedra	Loranthaceae	H	1625	11°33'26"	39°39'26"	MA123	1
Terminalia brownii Fresen.	Wyba	Combretaceae	T	1620	11°33'28"	39°39'25"	MA116	1
Thalictrum rhynchocarpum Quart.-Dill. & A.Rich.	Sire-Bizu	Ranunculaceae	H	1987	11°32'15"	39°36'02"	MA117	1
Tragia brevipes Pax	Aleblabit, Ablalit	Euphorbiaceae	H	1787	11°42'36"	39°39'22"	MA134	1
Trigonella foenum-graecum L.	Abish	Fabaceae	H	1783	11°43'06"	39°39'01"	MA118	1
Urtica simensis Hochst. ex A.Rich.	Sama	Urticaceae	H	1644	11°35'42"	39°39'33"	MA119	1
Vachellia amythethophylla (Steud. ex A.Rich.) Kyal. & Boatwr.	Lafto	Fabaceae	T	1930	11°44'28"	39°38'01"	MA152	2

Vachellia etbaica (Schweinf.) Kyal. & Boatwr.	Tikur Girar	Fabaceae	T	2040	11°47'12"	39°31'49"	MA153	2
Vachellia oerfota (Forssk.) Kyal. & Boatwr.	Ajo, Chelegama	Fabaceae	T	1860	11°32'37"	39°37'10"	MA01	1
Vachellia seyal (Delile) P.J.H.Hurter	Wacho Grar	Fabaceae	T	2017	11°47'26"	39°32'27"	MA03	1,2
Vachellia sieberiana (DC.) Kyal. & Boatwr.	Key Girar	Fabaceae	T	2226	11°47'00"	39°31'56"	MA154	2
Vachellia tortilis (Forssk.) Galasso & Banfi	Deweni Grar, Korera	Fabaceae	T	2138	11°44'09"	39°37'49"	MA04	1,2
Verbascum sinaiticum Benth.	Yahya Jero	Scrophulariaceae	H	1857	11°32'38"	39°37'09"	MA120	1
Verbena officinalis L.	Atuch	Verbenaceae	H	1869	11°32'25"	39°37'01"	MA121	1
Withania somnifera (L.) Dunal	Ede-buda/Gizawa	Solanaceae	S	1888	11°32'21"	39°36'27"	MA124	1
Zehneria scabra (Linn.f.) Sond.	Hareg resa/Etse Sabieq	Cucurbitaceae	H	1949	11°32'17"	39°36'02"	MA125	1
Zingiber officinale Roscoe	Zingibil	Zingiberaceae	H	1882	11°32'20"	39°36'57"	MA126	1
Ziziphus mauritiana Lam.	Kurkura	Rhamnaceae	T	1874	11°44'47"	39°37'09"	MA127	1
Ziziphus spina-christi (L.) Desf.	Kurkura, Geba	Rhamnaceae	T	2059	11°44'15"	39°38'07"	MA128	1,2

Key: **GF**, Growth form (Tree, T; Shrub, S; Herb, H; Climber, C); **1**, Plants reported for medicinal uses; **2**, Woody species documented in forest patche; **1,2**, Woody species from forest patches used for medicinal purposes.

Appendix 2: Synoptic cover abundance value of species in each community type

Species	**C1**	**C2**	**C3**	**C4**
Acacia decurrens (J.C.Wendl.) Willd.	0	0.27	0.02	0.32
Afrocarpus falcatus (Thunb.) C.N.Page	0	0.33	0.08	0
Albizia gummifera (J.F.Gmel.) C.A.Sm.	0	0	0	0.05
Allophylus abyssinicus (Hochst.) Radlk.	0.77	0.27	1.4	0.21
Arundo donax L.	0	0	0.04	0
Calotropis procera (Aiton) W.T.Aiton	0.08	0	0.02	0
Calpurnia aurea (Aiton) Benth.	0.08	0	0	0.05
Carissa spinarum L.	0	1.2	1	0.68
Casuarina equisetifolia L.	0	0	0	0.05
Celtis africana Burm.f.	0.08	0	0.02	0.16
Citrus aurantiifolia (Christm.) Swingle	0	0.27	0	0
Cordia africana Lam.	0	0	0.1	0
Croton macrostachyus Hochst. ex Delile	0	0	0.04	0.16
Dichrostachys cinerea (L.) Wight & Arn.	0.08	0	0	0
Dodonaea viscosa subsp. *angustifolia* (L.f.) J.G.West	2.92	4.93	4.21	3.47
Dombeya torrida (J.F.Gmel.) Bamps	0.08	0.07	0.21	0.05
Ehretia cymosa Thonn	0	0	0	0.21
Eucalyptus camaldulensis Dehnh.	1.77	0.67	0.62	4.89
Euclea racemosa L.	1.69	0.93	0.98	0.53
Euphorbia abyssinica J.F.Gmel.	0.77	0.2	1	0.42
Euphorbia tirucalli L.	0	0	0.04	0.16
Faidherbia albida (Delile) A.Chev.	0	0.07	0.08	0
Ficus capreifolia Delile	0.31	0	0	0
Ficus sur Forssk.	0	0	0.02	0
Ficus vasta Forssk.	0	0	0.12	0
Galiniera saxifraga (Hochst.) Bridson	0	0	0.02	0.05
Grevillea robusta A.Cunn. ex R.Br.	0	0	0.08	0
Grewia bicolor Juss.	0	0	0	0.05
Grewia ferruginea Hochst. ex A.Rich.	0.23	0.13	0.12	0.63
Gymnanthemum amygdalinum (Delile) Sch.Bip.	0.08	0.2	0.04	0
Gymnosporia senegalensis (Lam.) Loes.	0.08	0.4	0.54	0
Hesperocyparis lusitanica (Mill.) Bartel	1.92	0.07	0.04	0.53
Heteromorpha arborescens (Spreng.) Cham. & Schltdl.	0	0.2	0.15	0
Jacaranda mimosifolia D.Don	0	0	0	0.16

Juniperus procera Hochst. ex Endl.	7	0.67	1.38	0.21
Moringa oleifera Lam.	0	0	0.02	0
Myrsine africana L.	0	0.73	0.88	0.11
Olea europaea L. subsp. *cuspidata* (Wall. & G.Don) Cif.	2.54	2.13	5.52	3.16
Opuntia ficus-indica (L.) Mill.	0	0	0.06	0
Osyris lanceolata Hochst. & Steud.	1	0.53	0.71	0.68
Pittosporum viridiflorum Sims	1.62	5.07	2.9	0.53
Premna schimperi Engl.	0	0	0.17	0
Pterolobium stellatum (Forssk.) Brenan	0	0.47	0.21	0.16
Rhamnus prinoides L'Hér.	0	0	0.02	0
Rosa abyssinica R.Br. ex Lindl.	0	0	0.04	0
Schinus molle L.	0.23	0	0.08	0
Searsia glutinosa (Hochst. ex A.Rich.) Moffett	0.08	0.6	0.27	0
Searsia retinorrhoea (Steud. ex Oliv.) Moffett	0.77	3.2	0.44	0.26
Senegalia brevispica (Harms) Seigler & Ebinger	0	0.07	0.06	0
Vachellia amythethophylla (Steud. ex A.Rich.) Kyal. & Boatwr.	0	0	0	0.32
Vachellia etbaica (Schweinf.) Kyal. & Boatwr.	0.62	1.07	0.46	0.37
Vachellia seyal (Delile) P.J.H.Hurter	0.15	0	0.15	0
Vachellia sieberiana (DC.) Kyal. & Boatwr.	4.08	5.4	3.15	5.63
Vachellia tortilis (Forssk.) Galasso & Banfi	0	0	0.06	0
Ziziphus spina-christi (L.) Desf.	0	0.27	0.08	0.26

Key: C1 = Community type 1, C2= Community type 2, C3= Community type 3, C4= Community type 4

Appendix 3: Woody species list in Gerado forest patch with their respective Density (D), Frequency (F), Basal Area (BA), Relative Density (RD), Relative Frequency (RF), Relative Dominance (RDO), and Importance Value Index (IVI)

No.	Species name	D (h^{-1})	F	BA (m^2h^{-1})	RD	RF	RDO	IVI
1	*Acacia decurrens* (J.C.Wendl.) Willd.	6.40	11.63	0.18	0.28	1.24	0.69	2.21
2	*Afrocarpus falcatus* (Thunb.) C.N.Page	2.91	4.65	0.05	0.13	0.50	0.19	0.82
3	*Albizia gummifera* (J.F.Gmel.) C.A.Sm.	1.74	2.33	0.00	0.08	0.25	0.01	0.33
4	*Allophylus abyssinicus* (Hochst.) Radlk.	30.81	32.56	0.08	1.34	3.47	0.31	5.12
5	*Arundo donax* L.	4.65	2.33	0.00	0.20	0.25	0.02	0.47
6	*Calotropis procera* (Aiton) W.T.Aiton	5.81	4.65	0.00	0.25	0.50	0.02	0.77
7	*Calpurnia aurea* (Aiton) Benth.	3.49	4.65	0.01	0.15	0.50	0.04	0.69
8	*Carissa spinarum* L.	109.30	46.51	0.28	4.75	4.96	1.06	10.77
9	*Casuarina equisetifolia* L.	1.16	2.33	0.02	0.05	0.25	0.08	0.38
10	*Celtis africana* Burm.f.	6.98	9.30	0.06	0.30	0.99	0.25	1.54
11	*Citrus aurantiifolia* (Christm.) Swingle	1.74	2.33	0.01	0.08	0.25	0.02	0.35
12	*Cordia africana* Lam.	0.58	2.33	0.10	0.03	0.25	0.37	0.64
13	*Dodonaea viscosa* subsp. *angustifolia* (L.f.) J.G.West	519.19	95.35	1.28	22.57	10.17	4.91	37.65
14	*Dombeya torrida* (J.F.Gmel.) Bamps	5.81	6.98	0.02	0.25	0.74	0.06	1.06
15	*Ehretia cymosa* Thonn	6.40	4.65	0.07	0.28	0.50	0.29	1.06
16	*Eucalyptus camaldulensis* Dehnh.	81.98	30.23	0.93	3.56	3.23	3.57	10.36
17	*Euclea racemosa* L.	141.28	79.07	0.32	6.14	8.44	1.23	15.80
18	*Euphorbia abyssinica* J.F.Gmel.	66.28	18.60	0.90	2.88	1.99	3.46	8.32
19	*Euphorbia tirucalli* L.	7.56	2.33	0.02	0.33	0.25	0.09	0.67
20	*Faidherbia albida* (Delile) A.Chev.	2.91	4.65	0.01	0.13	0.50	0.03	0.65
21	*Ficus vasta* Forssk.	0.58	2.33	0.11	0.03	0.25	0.44	0.71
22	*Galiniera saxifraga* (Hochst.) Bridson	2.91	4.65	0.00	0.13	0.50	0.01	0.64
23	*Grevillea robusta* A.Cunn. ex R.Br.	1.74	2.33	0.10	0.08	0.25	0.38	0.70
24	*Grewia bicolor* Juss.	1.16	2.33	0.00	0.05	0.25	0.02	0.31
25	*Grewia ferruginea* Hochst. ex A.Rich.	8.14	13.95	0.02	0.35	1.49	0.06	1.91

26	*Gymnanthemum amygdalinum* (Delile) Sch.Bip.	5.23	4.65	0.01	0.23	0.50	0.05	0.77
27	*Gymnosporia senegalensis* (Lam.) Loes.	8.72	16.28	0.02	0.38	1.74	0.09	2.20
28	*Hesperocyparis lusitanica* (Mill.) Bartel	5.81	9.30	0.18	0.25	0.99	0.71	1.95
29	*Heteromorpha arborescens* (Spreng.) Cham. & Schltdl.	13.95	9.30	0.06	0.61	0.99	0.25	1.85
30	*Jacaranda mimosifolia* D.Don	2.91	2.33	0.06	0.13	0.25	0.23	0.60
31	*Juniperus procera* Hochst. ex Endl.	93.02	27.91	4.30	4.04	2.98	16.56	23.58
32	*Moringa oleifera* Lam.	2.91	2.33	0.02	0.13	0.25	0.09	0.46
33	*Myrsine africana* L.	62.21	27.91	0.09	2.70	2.98	0.35	6.03
34	*Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif.	394.77	83.72	5.42	17.16	8.93	20.86	46.95
35	*Opuntia ficus-indica* (L.) Mill.	1.16	2.33	0.03	0.05	0.25	0.10	0.40
36	*Osyris lanceolata* Hochst. & Steud.	45.93	37.21	0.12	2.00	3.97	0.45	6.41
37	*Pittosporum viridiflorum* Sims	275.00	90.70	4.96	11.95	9.68	19.08	40.71
38	*Premna schimperi* Engl.	5.81	2.33	0.01	0.25	0.25	0.03	0.53
39	*Pterolobium stellatum* (Forssk.) Brenan	24.42	23.26	0.07	1.06	2.48	0.27	3.81
40	*Rhamnus prinoides* L'Hér.	1.74	2.33	0.00	0.08	0.25	0.01	0.33
41	*Rosa abyssinica* R.Br. ex Lindl.	1.74	2.33	0.00	0.08	0.25	0.01	0.33
42	*Schinus molle* L.	2.33	2.33	0.02	0.10	0.25	0.08	0.43
43	*Searsia glutinosa* (Hochst. ex A.Rich.) Moffett	27.91	13.95	0.10	1.21	1.49	0.39	3.09
44	*Searsia retinorrhoea* (Steud. ex Oliv.) Moffett	111.63	48.84	0.41	4.85	5.21	1.57	11.63
45	*Senegalia brevispica* (Harms) Seigler & Ebinger	4.65	4.65	0.01	0.20	0.50	0.03	0.73
46	*Vachellia amythethophylla* (Steud. ex A.Rich.) Kyal. & Boatwr.	4.65	4.65	0.05	0.20	0.50	0.21	0.91
47	*Vachellia etbaica* (Schweinf.) Kyal. & Boatwr.	23.84	23.26	0.51	1.04	2.48	1.98	5.50
48	*Vachellia sieberiana* (DC.) Kyal. & Boatwr.	152.91	95.35	4.86	6.65	10.17	18.70	35.52
49	*Vachellia tortilis* (Forssk.) Galasso & Banfi	2.91	2.33	0.06	0.13	0.25	0.22	0.60
50	*Ziziphus spina-christi* (L.) Desf.	2.91	4.65	0.03	0.13	0.50	0.10	0.72

Appendix 4: Woody species list in Micha forest patch with their respective Density (D), Frequency (F), Basal Area (BA), Relative Density (RD), Relative Frequency (RF), Relative Dominance (RDO), and Importance Value Index (IVI)

No.	Species name	D (h^{-1})	F	BA (m^2h^{-1})	RD	RF	RDO	IVI
1	*Allophylus abyssinicus* (Hochst.) Radlk.	84.82	57.14	0.19	4.31	6.72	1.43	12.46
2	*Carissa spinarum* L.	28.57	21.43	0.06	1.45	2.52	0.42	4.39
3	*Croton macrostachyus* Hochst. ex Delile	3.57	3.57	0.02	0.18	0.42	0.12	0.72
4	*Dichrostachys cinerea* (L.) Wight & Arn.	2.68	3.57	0.01	0.14	0.42	0.09	0.64
5	*Dodonaea viscosa* subsp. *angustifolia* (L.f.) J.G.West	548.21	85.71	0.72	27.83	10.08	5.31	43.23
6	*Dombeya torrida* (J.F.Gmel.) Bamps	52.68	10.71	0.12	2.67	1.26	0.92	4.85
7	*Eucalyptus camaldulensis* Dehnh.	92.86	46.43	1.85	4.71	5.46	13.66	23.83
8	*Euclea racemosa* L.	57.14	42.86	0.10	2.90	5.04	0.75	8.69
9	*Euphorbia abyssinica* J.F.Gmel.	70.54	32.14	1.05	3.58	3.78	7.74	15.10
10	*Euphorbia tirucalli* L.	6.25	3.57	0.02	0.32	0.42	0.16	0.90
11	*Ficus capreifolia* Delile	1.79	3.57	0.01	0.09	0.42	0.04	0.55
12	*Ficus sur* Forssk.	0.89	3.57	0.00	0.05	0.42	0.01	0.48
13	*Grewia ferruginea* Hochst. ex A.Rich.	11.61	7.14	0.02	0.59	0.84	0.18	1.61
14	*Gymnanthemum amygdalinum* (Delile) Sch.Bip.	0.89	3.57	0.00	0.05	0.42	0.02	0.48
15	*Gymnosporia senegalensis* (Lam.) Loes.	34.82	32.14	0.06	1.77	3.78	0.41	5.96
16	*Hesperocyparis lusitanica* (Mill.) Bartel	27.68	28.57	0.53	1.41	3.36	3.90	8.67
17	*Heteromorpha arborescens* (Spreng.) Cham. & Schltdl.	11.61	3.57	0.03	0.59	0.42	0.25	1.26
18	*Juniperus procera* Hochst. ex Endl.	117.86	53.57	2.25	5.98	6.30	16.63	28.92
19	*Myrsine africana* L.	79.46	25.00	0.07	4.03	2.94	0.49	7.46
20	*Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif.	216.96	82.14	2.08	11.02	9.66	15.37	36.05
21	*Osyris lanceolata* Hochst. & Steud.	50.00	32.14	0.10	2.54	3.78	0.75	7.07
22	*Pittosporum viridiflorum* Sims	121.43	78.57	0.96	6.17	9.24	7.08	22.49
23	*Pterolobium stellatum* (Forssk.) Brenan	5.36	3.57	0.01	0.27	0.42	0.05	0.74
24	*Rosa abyssinica* R.Br. ex Lindl.	3.57	3.57	0.00	0.18	0.42	0.02	0.62

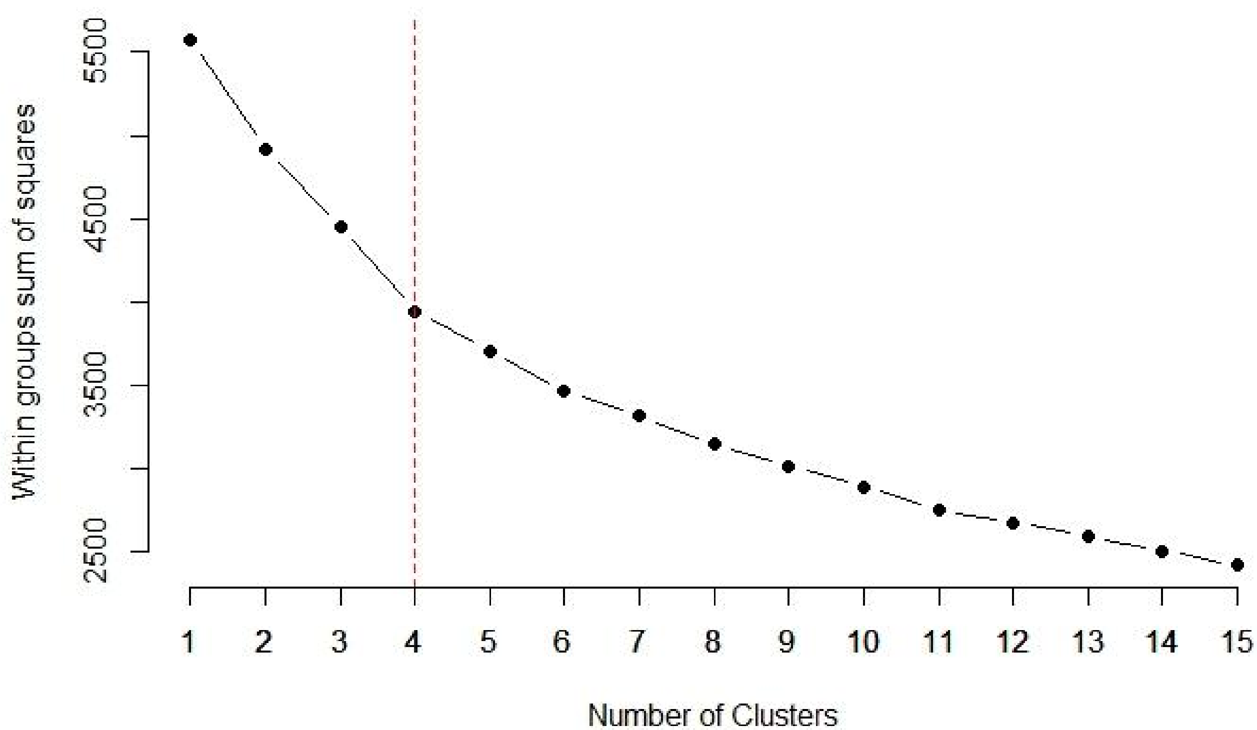
25	*Schinus molle* L.	1.79	3.57	0.01	0.09	0.42	0.04	0.55
26	*Searsia glutinosa* (Hochst. ex A.Rich.) Moffett	25.89	21.43	0.05	3.49	2.52	0.36	6.38
27	*Searsia retinorrhoea* (Steud. ex Oliv.) Moffett	57.14	35.71	0.07	2.90	4.20	0.51	7.61
28	*Senegalia brevispica* (Harms) Seigler & Ebinger	1.79	3.57	0.00	0.09	0.42	0.01	0.53
29	*Vachellia etbaica* (Schweinf.) Kyal. & Boatwr.	56.25	10.71	0.19	2.86	1.26	1.42	5.54
30	*Vachellia seyal* (Delile) P.J.H.Hurter	15.18	17.86	0.10	0.77	2.10	0.77	3.64
31	*Vachellia sieberiana* (DC.) Kyal. & Boatwr.	135.71	85.71	2.84	6.89	10.08	20.97	37.95
32	*Ziziphus spina-christi* (L.) Desf.	1.79	3.57	0.02	0.09	0.42	0.12	0.63

Appendix 5: Woody species list in Mekelet forest patch with their respective Density (D), Frequency (F), Basal Area (BA), Relative Density (RD), Relative Frequency (RF), Relative Dominance (RDO), and Importance Value Index (IVI)

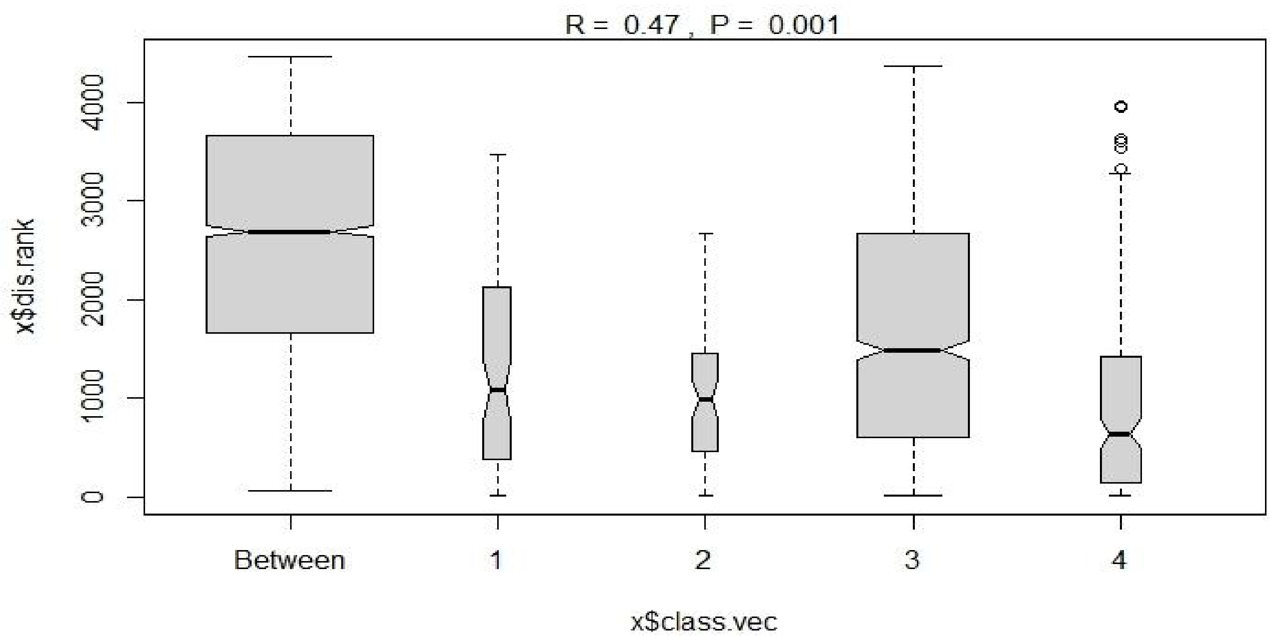
No.	**Species name**	**D (h^{-1})**	**F**	**BA (m^2h^{-1})**	**RD**	**RF**	**RDO**	**IVI**
1	*Allophylus abyssinicus* (Hochst.) Radlk.	47.92	54.17	0.12	2.27	8.18	0.75	11.20
2	*Carissa spinarum* L.	44.79	25.00	0.17	2.13	3.77	1.08	6.98
3	*Croton macrostachyus* Hochst. ex Delile	6.25	12.50	0.01	0.30	1.89	0.08	2.27
4	*Dodonaea viscosa* subsp. *angustifolia* (L.f.) J.G.West	833.33	91.67	1.79	39.55	13.84	11.59	64.97
5	*Eucalyptus camaldulensis* Dehnh.	141.67	50.00	1.21	6.72	7.55	7.86	22.13
6	*Euclea racemosa* L.	14.58	12.50	0.01	0.69	1.89	0.07	2.65
7	*Euphorbia abyssinica* J.F.Gmel.	17.71	12.50	0.06	0.84	1.89	0.36	3.09
8	*Faidherbia albida* (Delile) A.Chev.	2.08	4.17	0.00	0.10	0.63	0.02	0.75
9	*Grewia ferruginea* Hochst. ex A.Rich.	15.63	16.67	0.02	0.74	2.52	0.12	3.37
10	*Gymnanthemum amygdalinum* (Delile) Sch.Bip.	2.08	4.17	0.00	0.10	0.63	0.02	0.75
11	*Gymnosporia senegalensis* (Lam.) Loes.	26.04	25.00	0.06	1.24	3.77	0.36	5.37
12	*Juniperus procera* Hochst. ex Endl.	17.71	20.83	0.44	0.84	3.14	2.82	6.81
13	*Myrsine africana* L.	56.25	20.83	0.25	2.67	3.14	1.61	7.42
14	*Olea europaea* L. subsp. *cuspidata* (Wall. & G.Don) Cif.	304.17	83.33	3.57	14.43	12.58	23.12	50.14

15	*Osyris lanceolata* Hochst. & Steud.	59.38	41.67	0.11	2.82	6.29	0.74	9.85
16	*Pittosporum viridiflorum* Sims	15.63	29.17	0.18	0.74	4.40	1.18	6.32
17	*Premna schimperi* Engl.	11.46	12.50	0.06	0.54	1.89	0.41	2.84
18	*Pterolobium stellatum* (Forssk.) Brenan	3.13	4.17	0.01	0.15	0.63	0.04	0.82
19	*Searsia retinorrhoea* (Steud. ex Oliv.) Moffett	15.63	16.67	0.02	0.74	2.52	0.14	3.40
20	*Vachellia etbaica* (Schweinf.) Kyal. & Boatwr.	12.50	25.00	0.11	0.59	3.77	0.69	5.05
21	*Vachellia sieberiana* (DC.) Kyal. & Boatwr.	459.38	100.00	7.25	21.80	15.09	46.95	83.84

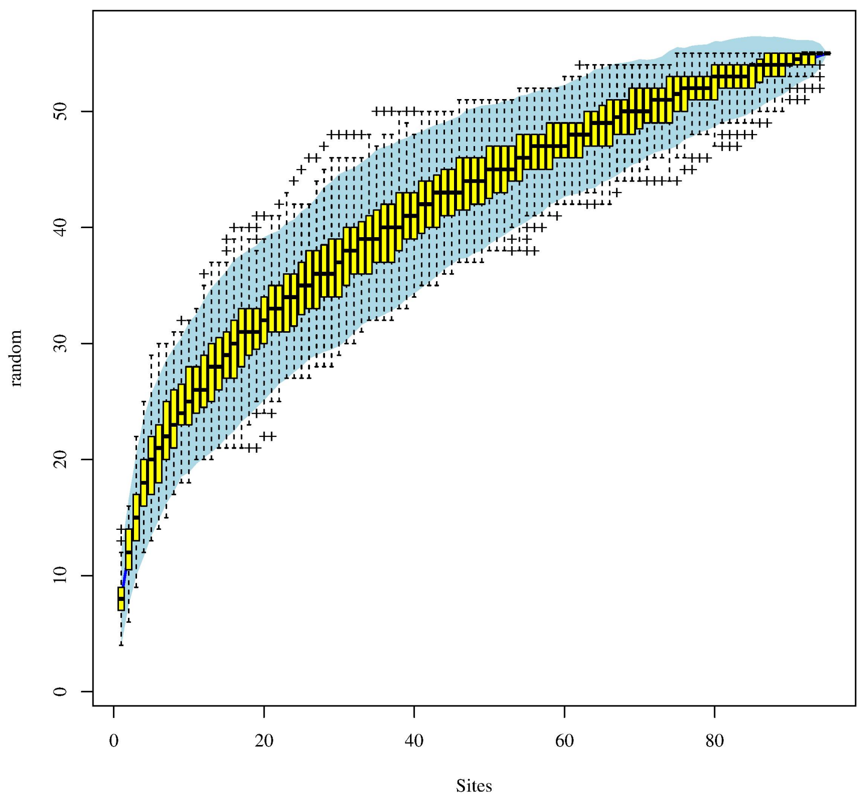
Appendix 6: Determining the optimal number of clusters in the studied forest patches



Appendix 7: MRPP significance test



Appendix 8: Species accumulation curve



Appendix 9: Minimum inhibitory concentration (MIC) standard

MIC Code	Concentration of standard (mg/ml)	Ciprofloxacin (µg/ml)
A	16	10
B	8	5
C	4	2.5
D	2	1.25
E	1	0.625
F	0.5	0.3125
G	0.25	0.15625
H	0.125	0.078125

Appendix 10: Minimum inhibitory concentration reading against gram positive bacterial strains

Bacterial Strain	Plant species & MIC Reading													
	Plant Sample Code	(S1) Solanum somalense Franchet. (Solanaceae)		(V2) Verbascum sinaiticum Benth. (Scrophulariaceae)		(R3) Rumex nervosus Vahl (Polygonaceae)		(W4) Withania somnifera (L.) Dunal (Solanaceae)		(C5) Calpurnia aurea (Ait.) Benth. (Fabaceae)		Ciprofloxacin		MBC
	Extraction Methods	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	
Gram-positive bacteria strains	*Staphylococcus aurous* (ATCC 25923)	C	A	C	A	B	A	D	D	B	>A	E	E	>A
		C	A	C	A	B	A	D	D	B	>A			
		B	B	C	A	B	A	D	D	B	>A			
		B	B	C	A	B	A	D	D	A	>A			
	Streptococcus agalactiae (ATCC 12386)	E	>A	C	A	>A	>A	C	C	>A	>A	E	E	>A
		E	>A	C	A	A	>A	C	C	>A	>A			
		D	>A	C	A	A	>A	D	C	A	>A			
		D	>A	C	A	A	>A	D	C	A	>A			
	Staphylococcus epidermidis (ATCC 12228)	A	>A	>A	>A	B	A	D	C	A	>A	E	E	>A
		A	>A	A	>A	B	A	D	C	A	>A			
		B	A	B	A	B	>A	C	C	A	>A			
		B	A	B	A	B	>A	C	C	A	>A			
	Enterococcus faecalis (ATCC 29212)	C	A	C	A	A	>A	C	C	>A	>A	D	D	>A
		C	A	C	A	A	>A	C	C	>A	>A			
		C	A	C	A	A	>A	C	C	A	>A			
		C	A	C	A	A	>A	D	C	A	>A			

EE = Ethanol extract; HE = Hydro xtract

Appendix 11: Minimum inhibitory concentration reading against gram negative bacterial strains

Bacterial Strain	Plant species & MIC Reading													
	Plant Sample Code	(S1) Solanum somalense Franchet. (Solanaceae),		(V2) Verbascum sinaiticum Benth. (Scrophulariaceae)		(R3) Rumex nervosus Vahl (Polygonaceae)		(W4) Withania somnifera (L.) Dunal (Solanaceae)		(C5) Calpurnia aurea (Ait.) Benth. (Fabaceae)		Ciprofloxacin		MBC (for all tested extracts)
	Extraction Methods	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	
Gram-negative bacteria strains	*Proteus mirabilis* (ATCC 35659)	C	>A	B	>A	A	>A	A	>A	B	>A	H	<H	>A
		B	>A	B	>A	A	>A	A	>A	B	>A			
		C	A	B	A	A	A	A	A	B	A			
		B	A	C	A	A	A	A	A	B	A			
	S. typhimurium (ATCC 13311)	C	A	C	B	A	B	B	B	B	A	F	E	>A
		C	A	C	B	A	B	B	B	B	A			
		C	A	C	B	A	B	B	B	B	A			
		C	A	C	B	A	B	B	B	A	A			
	Klebsiella pneumoniae (ATCC 700603)	C	A	C	A	B	B	B	A	D	A	F	E	>A
		C	A	C	A	B	B	B	A	C	A			
		D	A	C	B	B	A	B	A	D	B			
		D	A	B	B	B	A	B	A	C	B			
	Escherichia coli (ATCC 25922)	C	A	C	A	A	A	D	>A	D	A	<H	E	>A
		C	A	C	A	B	A	D	>A	D	A			
		C	B	C	A	A	A	D	A	D	A			
		D	B	B	A	B	A	D	A	D	A			
	Shigella flexneri (ATCC 12022)	B	B	C	C	A	A	C	C	A	>A	D	D	>A
		B	B	C	C	A	A	C	C	A	>A			
		B	B	D	C	A	A	C	D	B	>A			
		B	B	D	C	A	A	C	D	B	>A			

EE = Ethanol extract; HE = Hydro extract

Appendix 12: Minimum inhibitory concentration (MIC) mean and standard deviation of tested plants against gram positive bacterial strains

Bacterial Strain	Plant Sample Code	(S1) Solanum somalense Franchet. (Solanaceae)		(V2) Verbascum sinaiticum Benth. (Scrophulariaceae)		(R3) Rumex nervosus Vahl (Polygonaceae)		(W4) Withania somnifera (L.) Dunal (Solanaceae)		(C5) Calpurnia aurea (Ait.) Benth. (Fabaceae)		Ciprofloxacin	
	Extraction Methods	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE
Gram-positive bacteria strains	*Staphylococcus aurous* (ATCC 25923)	4	16	4	16	8	16	2	2	8	16	0.625	0.625
		4	16	4	16	8	16	2	2	8	16	0.625	0.625
		4	8	4	16	8	16	2	2	8	16	0.625	0.625
		8	8	4	16	8	16	2	2	16	16	0.625	0.625
	Mean	5	12	4	16	8	16	2	2	10	16	0.625	0.625
	Standard Deviation	2	4.62	0	0	0	0	0	0	4	0	0	0
	Streptococcus agalactiae (ATCC 12386)	1	16	4	16	16	16	4	4	16	16	0.625	0.625
		1	16	4	16	16	16	4	4	16	16	0.625	0.625
		2	16	4	16	16	16	2	4	16	16	0.625	0.625
		2	16	4	16	16	16	2	4	16	16	0.625	0.625
	Mean	1.5	16	4	16	16	16	3	4	16	16	0.625	0.625
	Standard Deviation	0.58	0	0	0	0	0	1.15	0	0	0	0	0
	Staphylococcus epidermidis (ATCC 12228)	16	16	16	16	8	16	2	4	16	16	0.625	0.625
		16	16	16	16	8	16	2	4	16	16	0.625	0.625
		8	16	8	16	8	16	4	4	16	16	0.625	0.625
		8	16	8	16	8	16	4	4	16	16	0.625	0.625
	Mean	12	16	12	16	8	16	3	4	16	16	0.625	0.625
	Standard Deviation	4.62	0	4.62	0	0	0	1.15	0	0	0	0	0
	Enterococcus faecalis (ATCC 29212)	4	16	4	16	16	16	4	4	16	16	1.25	1.25
		4	16	4	16	16	16	4	4	16	16	1.25	1.25
		4	16	4	16	16	16	4	4	16	16	1.25	1.25
		4	16	4	16	16	16	2	4	16	16	1.25	1.25

	Mean	4	16	4	16	16	16	3.5	4	16	16	1.25	1.25
	Standard Deviation	0	0	0	0	0	0	1	0	0	0	0	0

EE = Ethanol extract; HE = Hydro extract

Appendix 13: Minimum inhibitory concentration (MIC) mean and standard deviation of tested plants against gram negative positive bacterial strains

Bacterial Strain	Plant Sample Code	(S1) *Solanum somalense* Franchet. (Solanaceae),		(V2) *Verbascum sinaiticum* Benth. (Scrophulariaceae)		(R3) *Rumex nervosus* Vahl (Polygonaceae)		(W4) *Withania somnifera* (L.) Dunal (Solanaceae)		(C5) *Calpurnia aurea* (Ait.) Benth. (Fabaceae)		Ciprofloxacin	
	Extraction Methods	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE	EE	HE
Gram-negative bacteria strains	*Proteus mirabilis* (ATCC 35659)	4	16	8	16	16	16	16	16	8	16	0.078125	0.07813
		8	16	8	16	16	16	16	16	8	16	0.078125	0.07813
		4	16	8	16	16	16	16	16	8	16	0.078125	0.07813
		8	16	4	16	16	16	16	16	8	16	0.078125	0.07813
	Mean	6	16	7	16	16	16	16	16	8	16	0.078125	0.07813
	Standard Deviation	2.31	0	2	0	0	0	0	0	0	0	0	0
	S. typhimurium (ATCC 13311)	4	16	4	8	16	8	8	8	8	16	0.3125	0.625
		4	16	4	8	16	8	8	8	8	16	0.3125	0.625
		4	16	4	8	16	8	8	8	8	16	0.3125	0.625
		4	16	4	8	16	8	8	8	16	16	0.3125	0.625
	Mean	4	16	4	8	16	8	8	8	10	16	0.3125	0.625
	Standard Deviation	0	0	0	0	0	0	0	0	4	0	0	0
	Klebsiella pneumoniae (ATCC 700603)	4	16	4	16	8	8	8	16	2	16	0.3125	0.625
		4	16	4	16	8	8	8	16	4	16	0.3125	0.625
		2	16	4	8	8	16	8	16	2	8	0.3125	0.625
		2	16	8	8	8	16	8	16	4	8	0.3125	0.625

	Mean	3	16	5	12	8	12	8	16	3	12	0.3125	0.625
	Standard Deviation	1.15	0	2	4.62	0	4.6	0	0	1.15	4.62	0	0
	Escherichia coli (ATCC 25922)	4	16	4	16	16	16	2	16	2	16	0.078125	0.625
		4	16	4	16	8	16	2	16	2	16	0.078125	0.625
		4	8	4	16	16	16	2	16	2	16	0.078125	0.625
		2	8	8	16	8	16	2	16	2	16	0.078125	0.625
	Mean	3.5	12	5	16	12	16	2	16	2	16	0.078125	0.625
	Standard Deviation	1	4.62	2	0	4.62	0	0	0	0	0	0	0
	Shigella flexneri (ATCC 12022)	8	8	4	4	16	16	4	4	16	16	1.25	1.25
		8	8	4	4	16	16	4	4	16	16	1.25	1.25
		8	8	2	4	16	16	4	2	8	16	1.25	1.25
		8	8	2	4	16	16	4	2	8	16	1.25	1.25
	Mean	8	8	3	4	16	16	4	3	12	16	1.25	1.25
	Standard Deviation	0	0	1.15	0	0	0	0	1.15	4.62	0	0	0

EE = Ethanol extract; HE = Hydro extract

Appendix 14: Different photo shoots from the study sites

Forest patches and surrounding areas: a partial view (Photo courtesy: Mulugeta Alemu, 2021)

Anthropogenic disturbance in the study area: tree cutting, firewood collection and charcoal production (Photo courtesy: Mulugeta Alemu, 2023)

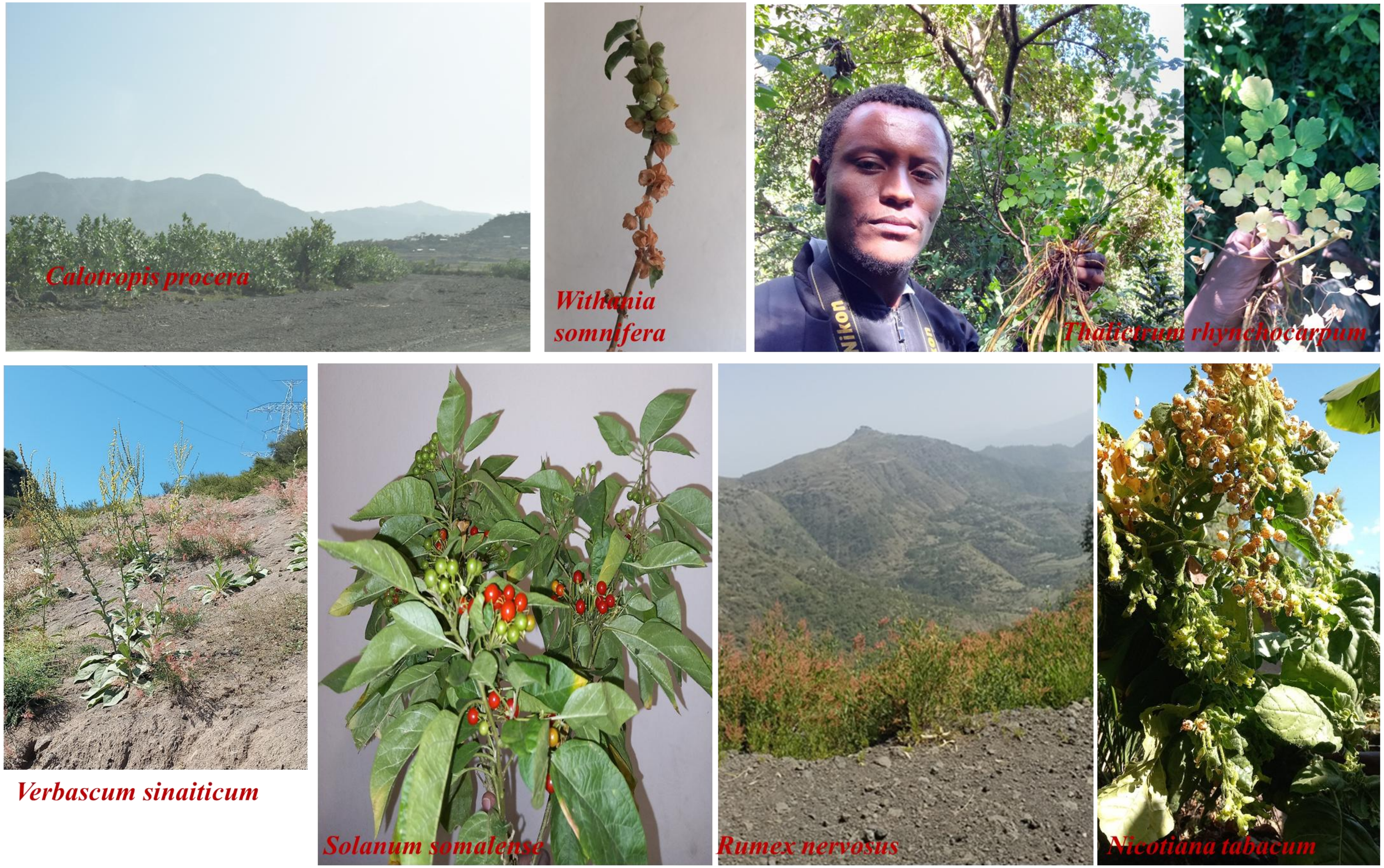



Sample photos of home-gardening in the study area (Photo courtesy: Mulugeta Alemu, 2022)

Sample photos of interviewing informants (Photo courtesy: Mulugeta Alemu, 2022)

Sample photos of FGD (Photo courtesy: Mulugeta Alemu, 2022)

Voucher specimen collection and drying plant materials at room temperature (Photo courtesy: Mulugeta Alemu, 2023)



Sample medicinal plants identified in the study area (Photo courtesy: Mulugeta Alemu, 2023)

Illustrative pictures for antibacterial activity and phytochemical screening of selected traditional medicinal plants (Photo courtesy: Mulugeta Alemu, 2023)

Appendix 15: Supportive letters

አዲስ አበባ ዩኒቨርሲቲ
የተፈጥሮ ሣይንስ ኮሌጅ
የዕፅዋት ባዮሎጂና የብዝሀ ሕይወት ማኔጅመንት ትምህርት ክፍል

ADDIS ABABA UNIVERSITY
College of Natural Sciences
Department of Plant Biology & Biodiversity Management

ቁጥር፡ DPBBM/CNCS/ /13/2021
ቀን፡ መጋቢት 7 ቀን 2013

ለሚመለከተው ሁሉ

ጉዳዩ፤ ትብብር ስለመጠየቅ ይሆናል

ከላይ በርዕሱ እንደተጠቀሰው ተማሪ ሙሉጌታ አለሙ በአዲስ አበባ ዩኒቨርሲቲ በድህረ ምረቃ ፕሮግራም በእፅዋት ባዮሎጂና ብዝሀ-ህይወት ማኔጅመንት ት/ክፍል (Plant Biology and Biodiversity Management) የPhD ተማሪ የሆነው ለጥናትና ምርምር (Research) ከመጋቢት 22 ቀን 2013 ዓ.ም ጀምሮ ላልተወሰነ ጊዜ ስለሚሠራ መ/ቤታችሁ ትብብር ታደርጉለት ዘንድ በትህትና እየጠየቅን ለሚደረግለት ትብብር ሁሉ ምስጋናችንን እናቀርባለን፡፡

ከሠላምታ ጋር

ቢቂላ ወርቅነህ (ዶ/ር)
የዕፅዋት ባዮሎጂና ብዝሃ ህይወት ማኔጅመንት ትምህርት ክፍል ኃላፊ

[illegible]

[illegible]

[illegible]

[illegible]

18/09/[illegible] ዓ.ም

Tel: +251- 0118959215; Email: pbbm_cns@aau.edu.et P.O. Box 3434; Addis Ababa, ETHIOPIA
In replying please Quote our Reference Number መልስ ሲጽፉልን እባክዎ የኛን ቁጥር ይጥቀሱልን

የኢትዮጵያ የሕብረተሰብ ጤና ኢንስቲትዩት
Ethiopian Public Health Institute
አዲስ አበባ-ኢትዮጵያ Addis Ababa, Ethiopia

ስልክ-Tel: +251 11 2133499, +251 11 2751522, ፋክስ Fax: +251 11 2758634,
የመ.ሳ.ቁ.-P.O. BOX: 1242/5654 e-mail: ephi@ethionet.et
www.ephi.gov.et

ቁጥር Ref No: ኢ.ሕ.ጤ.ኢ. 544/588
ቀን Date: 08 መጋቢት 2014

በአማራ ክልላዊ መንግስት
ለሰሜን ወሎ ዞን አስተዳደር
ወልዲያ

ጉዳዮ፡- የኢትኖቦታኒ ዳሰሳ ጥናት ለማመቻቸት ትብብርን በተመለከተ

የኢትዮጵያ የሕብረተሰብ ጤና ኢንስቲተትዩት ባልደረባ የሆኑት አቶ እዮብ ደበበ፣ አቶ ቢሆንኝ ሲሳይ፣ አቶ ሙሉጌታ አለሙ (ከአዲስ አበባ ዩኒቨርስቲ)፣ አቶ ደምመላሽ ሀ/ሚካኤል/ሾፌር/ ከመጋቢት 09/2014 ዓ.ም እስከ ሚያዚያ 09/2014 ዓ.ም በሰሌዳ ቁጥር ኮድ 4-15904 በሆነ መኪና በአማራ ክልላዊ መንግስት በሰሜን ወሎ ዞን በሀብሩ እና በጉባላፍቶ ወረዳዎች በሚገኙ የተለያዩ ቀበሌዎች በመዘዋወር የኢትኖቦታኒ የዳሰሳ ጥናት ለማመቻቸት ቀኝት ለማደርግ፣ መረጃዎችንና ናሙናዎችን ለመሰብሰብ ስለሚንቀሳቀሱ ለቡድኑ ተልእኮ መቃናትና ለሚካሄደው ጥናት ስኬት በሚሄዱበት ቦታ በቀበሌው አስተዳደር እና በሚመለከታቸው ሁሉ አስፈላጊውንና የተለመደውን መልካም ትብብር እንዲደረግላቸው በአክበሮት እንጠይቃለን፡፡

ከሠላምታ ጋር

ፍሬህይወት ተካ
ለ/ዘመም/ዳይሬክቶሬት
ተ/ዳይሬክከተር

ግልባጭ፡

- ለም/ዋና ዳይሬክቶሬት ፅ/ቤት
- ለአቶ እዮብ ደበበ
- አቶ ቢሆንኝ ሲሳይ
- አቶ ሙሉጌታ አለሙ
- አቶ ደምመላሽ ሀ/ሚካኤል/ሾፌር/

ኢ.ህ.ጤ.ኢ.

ቁጥር አሀ/ 7/2 /2013

ቀን 11/10/2013

ለሀብሩ ወረዳ ቀበሌ ጽ/ቤቶች

መርሳ፣

ጉዳዩ፡- ትብብር እንድደረግላቸው ይመለከታል፣

የኢትዮጵያ የህብረተሰብ ጤና ኢንስቲዩት ከአዲስ አበባ ዩኒቨርስቲ ጋር በመተባበር በወረዳችን በሚገኙ የተለያዩ ቀበሌዎች በመዘዋወር የኢትኖቦታኒ የዳሰሳ ጥናት ለማመቻቸት ቅኝት ለማድረግ መረጃዎችና ናሙና ለመሰብሰብ የተለያዩ ቀጣይ ጥናቶቻችን ለማመቻቸት እንድቻል የሁኔታዊ /መነሻ/ ጥናት ለማካሄድ ከሰኔ 07/2013 እስከ ሰኔ 25/2013 ዓ.ም በወረዳው የተለያዩ ቀበሌዎች ለሚንቀሳቀሱ ለቡድኑ ተልእኮ መቃናት እና ለሚካሄደው ጥናት ለኬት በሚሄዱበት ቦታ በቀበሌው አስተዳደር እና በሚመለከታቸው አካላት አስፈላጊውን ትብብር እንድደረግላቸው የኢትዮጵያ የህብረተሰብ ጤና ኢንስቲዩት በቁጥር ኢህጤኢ5.44/416 በቀን 04 ሰኔ 2013 በተፃፈ ደብዳቤ ጠይቀዋል::

በመሆኑም በያዙት የጊዜ ሰሌዳ ለማካኝነት በሚመጡበት ጊዜ አስፈላጊውን ትብብር በእናንተ በኩል እንድደረግላቸው እያሳሰብን ጥናቱን ለማድረግ የተላኩ ባለሙያዎች ስም ዝርዝር፡-

1. አቶ እዮብ ደበበ
2. አቶ ሳሙኤል ወ/ኪዳን
3. አቶ ሙሉጌታ አሰሙ
4. አቶ ወንድማገኝ ማሞ መሆናቸውን እንገልፃለን::

መልካም አስተዳደር እናሰፍን!!