An Ethnobotanical study of medicinal plants used by the local people of Assosa District, Benishangul Gumuz Regional State, Ethiopia

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Ethnobotany Research and Applications 29:66 (2024) - http://dx.doi.org/10.32859/era.29.66.1-29
Manuscript received: 10/10/2024 – Revised manuscript received: 05/12/2024 - Published: 08/12/2024

Research

Abstract

Background: Assosa District is home to variety of medicinal plants and associated indigenous knowledge where there was no previous study on medicinal plants. The study was aimed at documenting the medicinal plants and associated knowledge emphasizing structure and relationships among plants and humans.

Methods: About 58 regular and 18 healers were selected from nine sites based on availability of vegetation, healers and accessibility. Ethnobotanical techniques and indices were used for data collection and analysis supplemented by statistical tests, models and ordination methods. To display the structure of relationships among plants and humans, R Statistical Software was used.

Results: About 54 medicinal plants belonging to 49 genera and 33 families were identified. Family Solanaceae was with the highest number of species. Most of the medicinal plants were shrubs, followed by trees. Leaves were the most frequently used parts. About 64% of the remedies were prepared fresh. The highest fidelity level (100%) was recorded for hemorrhoids and gastric ulcers. The highest informant consensus factor values (0.96) were linked to diseases of the eye and adnexa. Deforestation and urbanization were among the major threats to conservation. Significant associations among medicinal plant knowledge and sociodemographic factors observed.

Conclusion: There is rich medicinal plant diversity and associated indigenous knowledge demanding ex-situ and in-situ conservation. Phytochemical profiling of high-ranking medicinal plants for cupping therapy and other health problems is needed. Ordination methods are good representations of the structure of relationships among plants and humans.

Keywords: Assossa, cupping therapy, Ethiopia, Ethnobotany, Herbs, Indigenous

Background

Plants have historically served as sources of medicinal compounds for the treatment of various diseases across the globe, a practice that dates back to ancient times (Maiyo *et al*. 2024; Zemede *et al*. 2024). This tradition is particularly significant in developing countries, including Ethiopia. Medicinal plants, defined as those possessing bioactive substances with pharmacological effects on the human body or its organs, are utilized for therapeutic purposes (Yudharaj *et al.* 2016). These plants offer dual benefits: they provide nutritional value while also serving medicinal and ritual purposes (Flatie *et al.* 2009). Their role remains critical in the healthcare systems of many local communities, serving as the primary source of medicine for the majority of rural populations.

There are 6027 species of higher plants in the Ethiopian Flora, of which 27 of them are subspecies (Kelbessa & Demissew 2014). This indicates that Ethiopia is a country characterized by a wide range of climate and ecological conditions and possesses an enormous diversity of flora and fauna having medicinal importance in health care system based on local indigenous knowledge (IK) rural and urban. As a result, more than 80% of the Ethiopian population has still relied on those medicinal plants for their health treatment (Zemede *et al*. 2024). However, it does not mean that 80% of the population lives in rural areas. Population projection from Ethiopian Statistical Service (ESS 2013) showed that 76.84% the population lives in rural areas.

As defined in many publications, IK refers to the accumulation of knowledge, rules, standards, skills, and mental sets, which are possessed by local people in a particular area (Zerabruk & Yirga 2011). Thus, IK is the result of many generations' long years' experiences, careful observations, trial and error experiments. Indigenous knowledge develops and changes with time and space with the change of resources and culture (Fenetahun & Eshetu 2016).

Knowledgeable individuals in traditional medicine often refrain from sharing their expertise with the broader community where they reside. Instead, they prefer to keep this knowledge confined within their family circle. Traditional healers typically lack written records, relying instead on oral transmission of knowledge from one generation to the next. As a result, there is limited access to documented information and records about medicinal plants (Cunningham, 2001). Furthermore, Ethiopia's diverse cultures, beliefs, languages, and geographical settings contribute to a rich yet fragmented repository of medicinal plant knowledge, with the potential for a wide variety of uses and practices.

In the Assossa District, specifically, and the Benishangul-Gumuz Region of Ethiopia, in general, the lack of documentation and the secrecy surrounding traditional knowledge are significant contributors to the erosion and depletion of this valuable indigenous knowledge. Additionally, high rates of deforestation driven by agricultural expansion, investment projects, and inadequate forest conservation practices are threatening the survival of medicinal plants in the area. A report by Awas (2007) documented 185 plant species used in the daily lives of the Berta and Gumuz people in the region. Of these, approximately 30% are cultivated, while 70% are harvested from the wild, reflecting an unsustainable approach to resource use.

Ethnobotanical studies are essential for documenting the interaction between medicinal plants and human societies (Tadesse *et al.* 2005). While significant research has been conducted on ethnobotany in various parts of Ethiopia in recent years, little to no studies have focused on the Benishangul-Gumuz Regional State, particularly in the Assossa District. This lack of research may be attributed to the region's remote location bordering Sudan to the west, limited road accessibility, and hot climatic conditions, despite its richness in ethnic, linguistic, cultural, and vegetation diversity. To the best of our knowledge, no studies have been conducted on the ethnobotanical practices specific to this district. This gap inspired the researchers to undertake this study with specific objectives: to identify plant species with medicinal properties used for treating human diseases and in cupping therapy; to determine the composition, growth forms, and habitats of these medicinal plants; to examine the medicinal parts of plants, their methods of preparation, additives, dosage, marketability, and applications; to rank these plant species based on their importance and potential for treating diseases using various ranking methods; and to identify the primary threats to medicinal plants in the area.

Materials and Methods

Description of the study area

Geographical location

Benishangul-Gumuz Regional State, situated in northwestern Ethiopia, is one of the country's regional states. It is divided into three administrative zones: Assosa, Kamashi, and Metekel. The region derives its name from two indigenous ethnic groups, the Berta (also known as Benishangul) and the Gumuz. Assosa serves as the administrative capital of the region. Geographically, it shares borders with the Amhara Regional State to the north and northeast, Oromia Regional State to the south and southeast, and the Republic of Sudan to the west (UNICEF, 2019) (Figure 1).

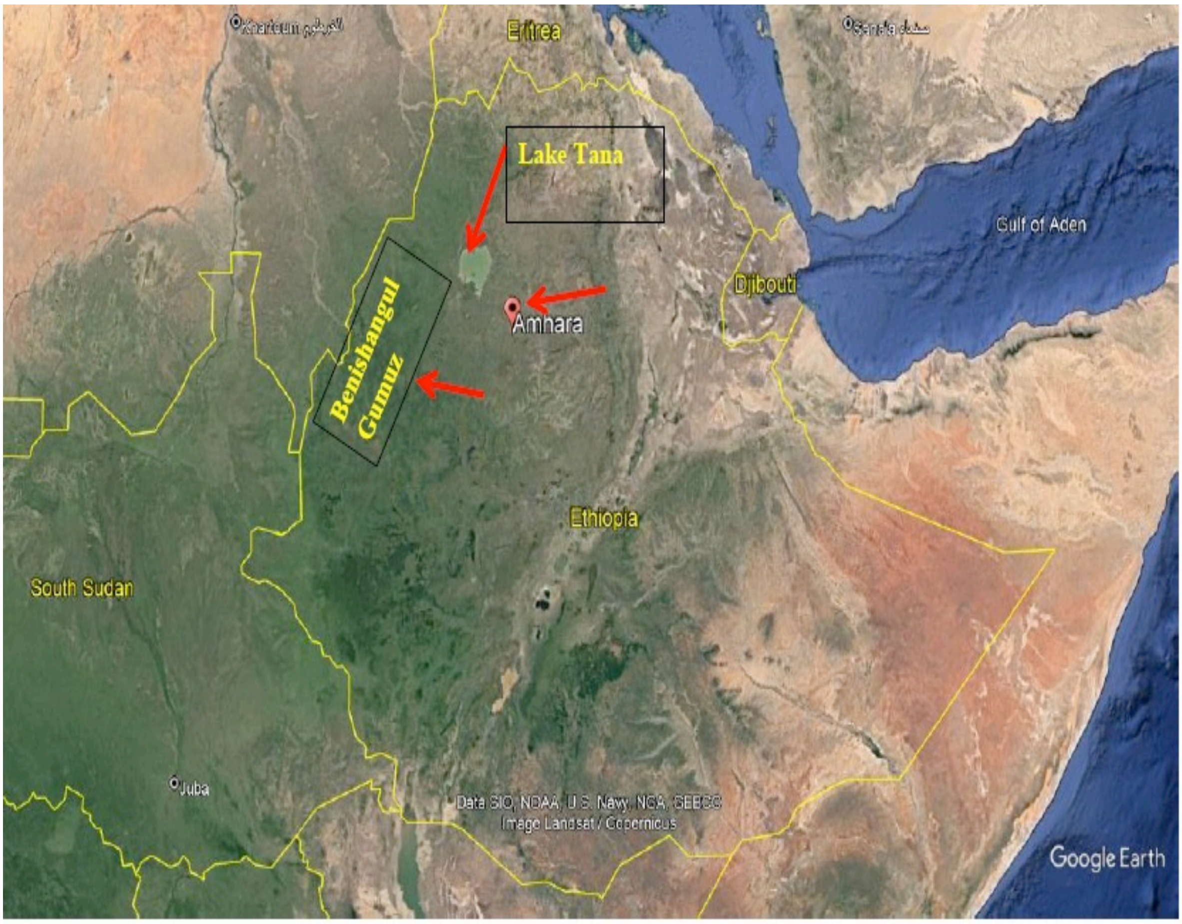

Figure 1. Google Earth image of the study area (Source: Google Earth Pro Landsat image: 19/01/2024).

The study was conducted in Assosa district, Assosa Zone, Benishangul Gumuz Regional State located in western Ethiopia at a longitude and latitude of 34°32' 45.2" E and 10°02' 44.4"N, with an average elevation of 1541 meters above sea level. The town is also situated at 667 km far away from Addis Ababa, the capital of Ethiopia (Figure 2).

Climate: rainfall and temperature

In Assosa, the climate features a warm and overcast wet season and a hot, partly cloudy dry season. The recorded minimum and maximum temperatures are 12.6°C and 32.2°C, respectively, with fourteen-years average temperature of 21.3°C based on Meteorological data collected over 14 years from Assosa station of the Meteorology Service Agency of Ethiopia. As shown in Figure 3, the rainfall pattern in the district is unimodal. The rainy season, locally known as *Keremt*, lasts from June to September, peaking in August and September. The highest average monthly rainfall, 246.5 mm, occurs in August, while January sees the lowest, at just 0.1 mm. The hottest months are March to November, with the maximum mean temperature of 32.2°C recorded in March and April. The coldest months are November to January, with the lowest temperature of 12.6°C observed in November and December. Overall, the district's mean annual temperature is 21.3°C, and the mean annual rainfall is 1,236 mm (Figure 3).

Population and healthcare facilities

According to the current population projection of Ethiopia based on (ESS 2013; CSA 2008), the Benishangul-Gumuz Region has a total population of 1,187,997 consisting of 602,999 men and 584,998 women. In Ethiopia, the hospital-to-worker ratio stands at 1:564,173, reflecting the underdeveloped state of the country's healthcare system (Firehiwot Berhane *et al.* 2015). This significant shortage of healthcare workers is one of the key factors driving reliance on medicinal plants, particularly in the country's remote areas. In Benishangul-Gumuz Region, population-Medical Doctor Ratio is also low when compared with other regions of the country next to Gambella (Adugna 2014). A population-hospital bed ratio of Bensangul Gumuz Region is the lowest one. According to the report of UNICEF (2019), the under-five mortality rate in Benishangul-Gumuz was the highest (98/1,000 live births) in the country next to Afar region. According to the respondents of the study area, hepatitis, hemorrhoids, snake bit, and asthma are the highest frequently occurring diseases.

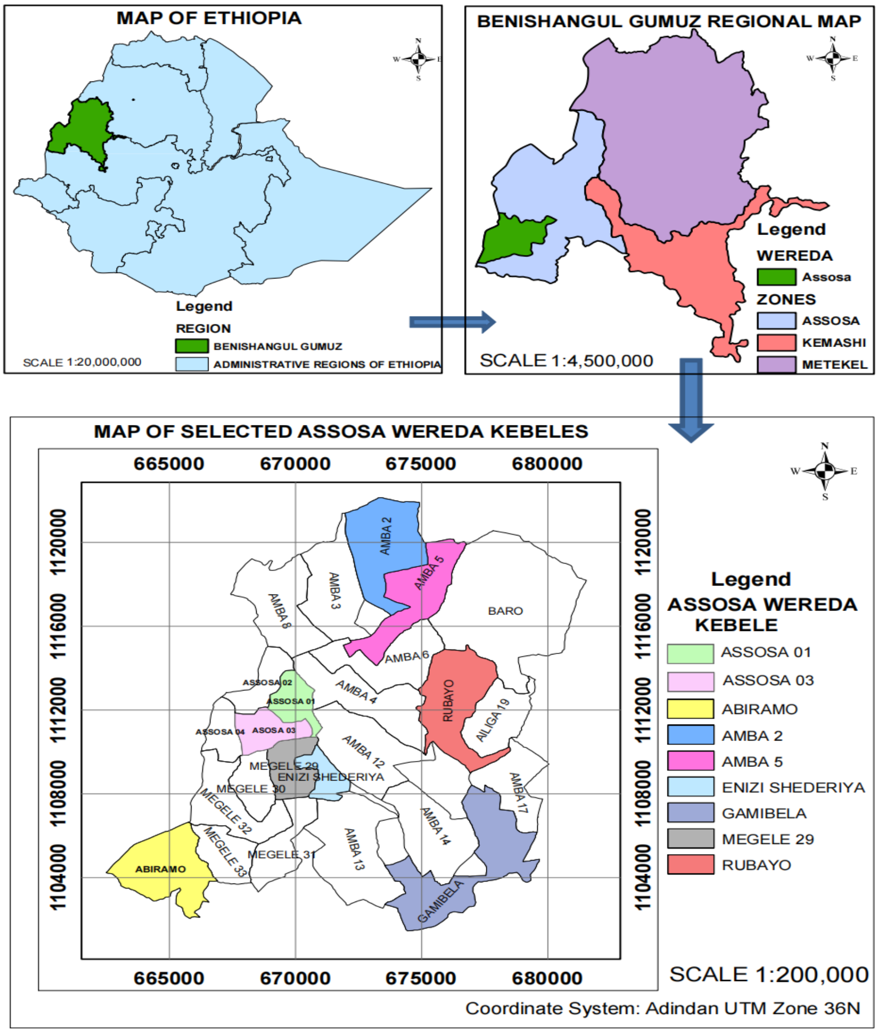

Figure 2. Location map of the study area

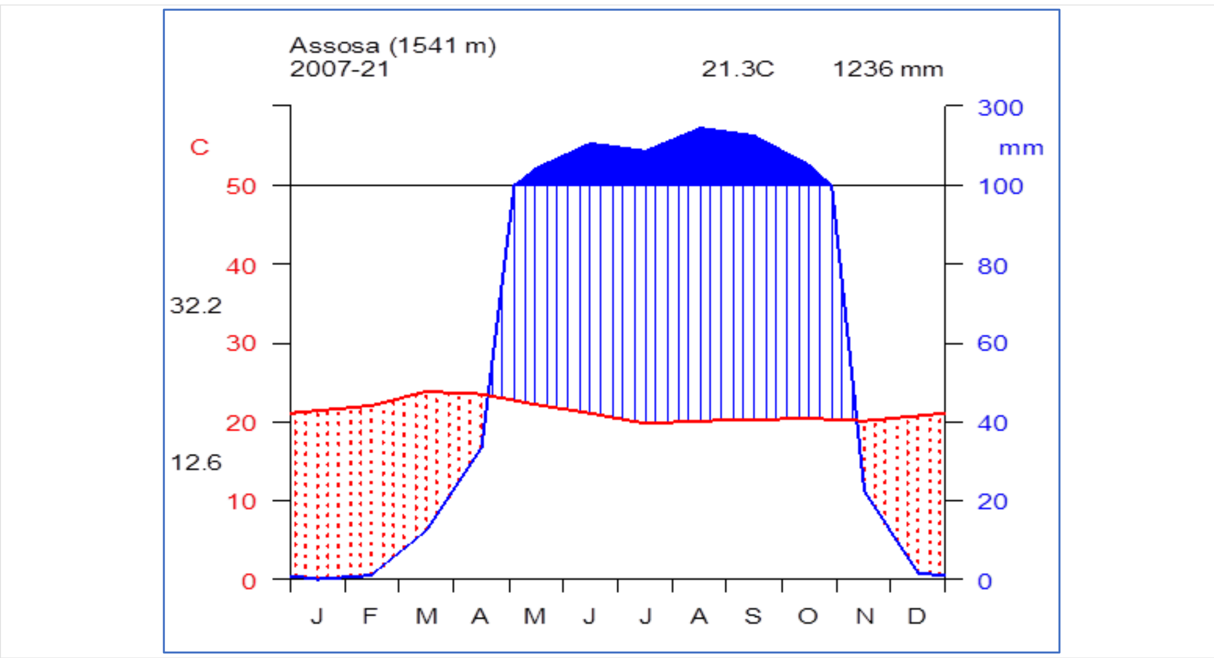

Figure 3. Climate Diagram of the study area

Vegetation

The Benishangul Gumuz Region, located in western Ethiopia, is renowned for its diverse vegetation. As noted by Awas *et al.* (2007), the region's flora is classified as "undifferentiated woodlands (Ethiopian type)," showcasing unique and partially distinct plant species. Much of this vegetation remains relatively intact, dominated by broadleaved deciduous trees. Remarkably, six plant species—*Acalypha bipartita*, *Dalbergia boehmii*, *Dorstenia benguellensis*, *Hyparrhenia bracteata*, *Ochna leptoclada*, and *Scleria greigiifolia*—were recorded in the region as new additions to Ethiopia's flora.

Abera and Yasin (2018) further describe the region as being characterized by relatively tall trees, with at least 20% canopy coverage, even in areas that include open spaces and felled lands awaiting regeneration. Prominent tree species in the area include Combretum species, Terminalia species, *Cordia africana*, *Adonsonia digitata*, and *Tamarindus indica*. Additionally, the landscape features small trees, bushes and shrubs unique to the region, often interspersed with grasses. The grasses, the predominant natural vegetation, are vital for grazing and browsing animals.

Reconnaissance survey and study site selection

Reconnaissance survey of the study area was made for the first two weeks of October, 2021. General information and intimate knowledge of the area were obtained during the survey. This survey was also used to familiarize with the study area. During this survey, the district administrative bodies and agricultural experts were consulted for selecting the study sites (Kebeles are the smallest governmental administrative units) for their availability of vegetation, traditional healers, and accessibility (security, road accessibility also). A total of nine (two from urban and seven from rural) kebeles were selected from Asossa district. Those selected kebeles were Assosa 01 (1579 m.a.s.l.), Rubayo (1573), Amba 2 (1561), Gambella (1488), Amba 5 (1571), Abrehamo (1497), Assosa 03 (1579), Megele 29 (1522) and Enzi shederya (1572) kebeles.

Respondent selection and sampling design

Regular informants and knowledgeable traditional healers (key informants) of the kebeles were selected using random and purposive sampling techniques, respectively, following Martin (1995). Those general informants were ordinary people of the district, were interviewed as respondents to gather additional data and check the transfer of indigenous knowledge within the people. Interviews and Focal Group Discussions (FGD) were held with the key informants who are practitioners/experts/ in the study area. However, before selecting the key informants, elders, development agents (DA) and administrative bodies of the kebeles were consulted. Their recommendations were useful on how to locate and approach the local traditional healers and carryout the field-guided plant survey. Legal supportive letter from Assosa district for all studied kebele was written in order to contact with those key informants. Some of the key informants are legal experts (known and register in government office) and others are non-legalized.

Accordingly, ethnobotanical data were collected from 76 informants (54 males and 22 females). From the total, 18 were key informants who were selected using purposive sampling technique, while the other 58 regular informants who were selected using random sampling technique. In terms of the religious of those informants, 47%, 28%, and 25% were Orthodox, Protestant, and Muslim, respectively. The age of respondents ranged from 25-86 years old of which 36 informants had less than 40 years old and the remaining 40 informants were greater than or equality to 40 years old.

Ethnobotanical data collection

The ethnobotanical data were collected by using different data collection methods as described in detail here under.

Semi-structured interview: It is a type of interview that contains a checklist of topics and questions that we want to cover, like the plant name, use, and way of preparation, application, additives used and other related topics. The local name of medicinal plants, part used, disease treated, dosage, methods of preparation, additive used for preparation, route of administration, use of the plant other than medicinal uses and conservation methods were recorded (Figure 4). The questions were determined beforehand, and others arise during the conversation (Martin 1995). Following this principle, general information and ethnobotanical data were collected using interviews. Semi-structured interviews were conducted with a single person face to face interview at a time. The questions for the interview were both open-and close-ended. The informants were interviewed individually using their local languages (Berta, Shinasha, Afaan Oromo and Amharic) for their understandability of the questions raised by the researcher and translators of the language. The information was carefully recorded during the interview with participants using notebooks and video tape).

Focus group discussion (FGD): Participants can build consensus by discussing an issue among themselves and agreeing on an answer or agreeing to disagree (Martin 1995). Discussions were made with two groups of 18 participants, who were drawn from each selected kebeles (about two individuals per kebele). This is because FGD is a highly effective technique to get detail information about the acceptability of the medicinal plant among the community, to give ranks of the most preferred medicinal plants, to distinguish the plants which are at risk (status), as well as the conservation practices and threats to medicinal plant. Alexiades (1996) also stated as FGDs can produce a wealth of data and lead to the discovery of new topics and questions. The FGDs were, thus, made to gain detailed information on plant knowledge and prove the reliability of the data collected through semi-structured interviews.

Figure 4 Photo showing the interview made with one of the key informants

Guided-field-walk*:* To create an opportunity to make note of the habit, habitat, appearance of the medicinal plant species, a guided field-walk was conducted with practitioners throughout the habitats where they are found (Figure 5). Traditional healers, who helped during the guided field walk, played a vital role in identifying the medicinal plant encountered in the field by providing its local (vernacular) name and medicinal use (Kassa 2009).

Figure 5. Guided field walk

Market survey: In any site and town, markets are places where medicinal plants, fruits, and other plant products are sold. These markets are rich sources of ethnobotanical information (Alexiades 1996). Following Martin (1995), many medicinal plants, ornamental, wild foods and other products have a strictly regional value which can only be discovered by talking with producers, sellers and consumers. Hence, the information like observing and recording the plant species, the way of preparation of the plant, threated plants also recorded; such like information was drawn from the local community (consumer) of the study area with greater care and confidentiality (Figure 6). The multipurpose roles of some medicinal plants were observed during market surveys. Hence, observations on public cultural celebrations and rituals, market surveys, individual or group activities, and the way the people of the study area react to their environment were carefully made, recorded, and interpreted during the study.

Figure 6. Market surveys made during data collection period for the study

Questionnaire-based survey: In addition to the above ethnobotanical data collection questioners were also used as a data collecting tools. In order to understand the attitude/perception/, acceptance of traditional healing method of the community we tried to distribute the questioner to regular informants.

Plant specimen collection and identification

After recording the ethnobotanical information, voucher specimens of medicinal plants were collected during guided field walks with the key informants. The specimens were numbered, pressed, dried, and taken to Debre Birhan University for identification. Identification of specimens was carried out by using the Flora of Ethiopia and Eritrea (Edwards *et al*. 2000, Edwards *et al*. 1997, Edwards *et al*. 1995, Hedberg *et al*. 2009, Hedberg *et al*.2006, Hedberg *et al*. 2003, Hedberg & Edward 1989, Phillips1995, Tadesse 2004). Further identification was also made in the National Herbarium of Addis Ababa University (ETH) with the help of an expert and authenticated specimens. Finally, the voucher specimens and their copies were deposited at Herbarium of Addis Ababa and Debre Birhan Universities, respectively.

Plant use classification

In this survey, eleven ailment categories were frequently mentioned and were treated with a wide range of medicinal plants. According to International Classification of Diseases (ICD)-10, 16 different disease categories were listed. The ailments that were addressed by the reported medicinal plants were categorized according to the ICD-10 version 2016. The classification results were utilized in the calculation of Informant Consensus Factor (ICF).

Ethnobotanical values and calculations

The data were analyzed and described using descriptive statistical methods such as frequency, percentage, tables, graphs, and figures via applying Microsoft Excel Spreadsheet 2010 and SPSS. Moreover, some qualitative data were analyzed using content analysis, narrating via drawing sub-contents. Ordination methods were used in R Statistical Software to complement the ethnobotanical indices.

Comparative analysis

Ranking methods such as preference ranking, direct matrix ranking, informant consensus factors, and fidelity level measurements were applied to determine the degree of importance of medicinal plants as described here under in detail.

Preference ranking

Preference ranking was conducted following Martin (1995) for the six most important medicinal plants selected by the informants used to treat hemorrhoids, the most frequently reported disease. Eight purposively selected key informants participated to identify the best preferred medicinal plant species for treating hemorrhoids by giving the medicinal plants for those informants by asking them to arrange the medicinal plants based on their effectiveness by assigning the highest value (6) for the most preferred plant species and the lowest value (1) for the least preferred ones. Finally, these values were summed up and ranked for each plant species. Thus, this method is used to know the most preferred medicinal plants to treat the disease hemorrhoid, which was frequently reported by the key informants.

Direct matrix ranking
Direct matrix ranking exercise was done following Martin (1995) to compare the multipurpose use of a given species. Based on the information gathered from the informants, six multipurpose tree species were selected out of the total medicinal plants and six use categories of these plants were also listed for eight randomly selected general informants to assign use values to each species. The six uses included medicinal use, food, firewood, construction, charcoal, and furniture. Accordingly, each chosen informant was asked to assign five use values (5 to the best use; 4, very good; 3, good; 2, less used; 1, least used, and 0, not used). The values (average score) of each species were summed up and ranked. Similarly, the values of each use type were calculated and summed up for ranking each use type. Direct matrix ranking is used to compare multipurpose uses of a given plant species based on information gathered from participants. Preference ranking is based on a single dimension, whereas direct matrix ranking draws explicitly upon multiple dimensions (Martin 1995).

Fidelity level
Fidelity level (FL) values were used to estimate the relative healing potential of each medicinal plant based on the proportion of informants who agreed on its use against a given ailment category (Friedman *et al*. 1986). Fidelity level was calculated as:

$$FL = \frac{Ip}{Iu} x100$$

Where - Ip is the number of informants who independently cited the importance of a species for treating a particular disease. Iu is the total number of informants who reported the plant for any given disease.

Informant consensus factor (ICF)
The agreement of the informants on the reported use of medicinal plants to cure human disease will be tested by calculating the ICF (Alexaides 1996). Thus, the ICF was calculated as:

$$ICF = \frac{nur - nt}{nur - 1}$$

Where $\mathbf{n_{ur}}$ = number of use citations in each category of disease. $\mathbf{n_t}$ = total number of medicinal plant species used (Heinrich *et al*. 1998, Andrade-Ceto & Heinrich, 2011).

Results

Socio-demographic characteristics of respondents
In the current study, more number of medicinal plants was reported by men than that reported by women. The participants, 71% male and 29% female, are participating in this study. When we see the religious category of those informants, 47% (orthodox), 28% (Protestant), and 25% (Muslim) are participating in this study (Table 1).

Table 1. Socio-demography of the informants

Characteristics	Informant detail	Frequency	Percentage
Gender	Male	54	71.00%
	Female	22	29.00%
Religion	Orthodox	36	47.00%
	Protestant	21	28.00%
	Muslim	19	25.00%
Age	< 40	36	47.00%
	≥40	40	53.00%
Informant categories	Regular informants	66	87.00%
	Key informants	10	13.00%
Educational status	Modern education	60	79.00%
	Church education	5	7.00%
	Uneducated	11	14.00%
	Total	76	100.00%

Statistical analysis of sociodemographic characteristics and use value variables

Ordination methods were used to stablish relationships between humans and medicinal plants. Constrained ordination was performed using Deterended Correspondence Analysis (DECORANA) for sociodemographic variables and Canonical Correspondence Analysis (CCA) for use value variables (Tables 2, 3).

Table 2. Significance of sociodemographic variables (P ≤ 0.05) for knowledge differences

Variables	Df	AIC	F	FPr(>F)	Codes	Significance
Age	1	324.63	3.9598	0.005	**	yes
Informant category	1	325.70	2.8678	0.005	**	yes
Religion	1	326.12	2.4379	0.005	**	yes
Education	1	327.25	1.3139	0.135		no
Gender	1	327.32	1.2498	0.175		no

Key: - Significance codes: 0”***”0.001”**”0.01”*”0.05”.”0.01”1

Table 3. Anova.cca of use value variables (P ≤ 0.05) for potential destructive impacts

UV variables	Df	SumOfsqs	Meansqs	F.Model	R^2	Pr(>F)	Codes	Significance
Firewood	1	0.7968	0.79676	2.0090	0.02660	0.020	*	yes
Medicinal	1	0.4470	0.44703	1.12717	0.01492	0.280		no
Construction	1	0.3820	0.38203	0.96328	0.01275	0.390		no
Furniture	1	0.2938	0.29379	0.74077	0.00981	0.770		no
Charcoal	1	0.3238	0.32379	0.81644	0.01081	0.730		no
Food	1	0.3453	0.34531	0.87069	0.01153	0.590		no
Residuals	69	27.3650	0.39659		0.91358			
Total	75	29.9537			1.00000			

Key: - Significance codes: 0”***”0.001”**”0.01”*”0.05”.”0.01”1

Pearson correlation

Correlation coefficients of medicinal plant knowledge differences among informants based on sociodemographic variables at P<0.05 showed that there was strong correlation between age and gender (r=0.3600, p=0.0014), age and informant category (r=0.7200, p=0.0000), age and religion (r= 0.8100, p=0.0000), gender and informant category (r= 0.0016, p=0.0016), gender and religion (r= 0.2600, p=0.0260), informant category and education (r= 0.30, p=0.0079) and informant category and religion (r= 0.500, p=0.0000) as indicated in Table 4.

Table 4. Correlation coefficients of medicinal plant knowledge differences among informants

Variables	**Age**	**Gender**	**Informant category**	**Education**	**Religion**
Age	1.0000				
≥40, n=40					
<40, n=36					
Gender		1.0000			
Male, n=44	0.3600				
Female, n=32	p=0.0014*				
Informant category			1.0000		
Regular informants, n=58	0.7200	0.3600			
Healers, n=18	P=0.0000*	P=0.0016*			
Education				1.0000	
Modern education, n=60	0.0800	0.0800	0.300		
Church education, n=5	P=0.5042	P=0.4863	P=0.0079*		
Uneducated, n= 11					
Religion					1.0000
Orthodox, n=36	0.8100	0.2600	0.500	-0.5000	
Protestant, n=21	P=0.0000*	P=0.0260*	P=0.0000*	P=0.6770	
Muslim, n=19					
Total (N)=76					

*=Indicates the correlation coefficient (r) is statistically significant at P<0.05; Bold=Indicates sociodemographic variables

Diversity and habits of the medicinal plants

A total of 54 medicinal plants belonging to 49 genera and 33 families were identified from the study area. Family Solanaceae was the most dominant medicinal plant in the study area, and contributed high number of medicinal plant species, which might be because of the hottest climatic and/or the lowest altitudinal conditions in the majority of Assosa District (Table 5). This also indicates that the area consisted of a considerable diversity of plant species.

Table 5. Taxonomic diversity of medicinal plants in the study area

Family	Genera		Species	
	Frequency	Percentage	Frequency	Percentage
Solanaceae	3	6.12%	5	9.26%
Apiaceae	3	6.12%	3	5.56%
Asteraceae	2	4.08%	3	5.56%
Cucurbitaceae	3	6.12%	3	5.56%
Fabaceae	2	4.08%	3	5.56%
Malvaceae	3	6.12%	3	5.56%
Myrtaceae	3	6.12%	3	5.56%
Rutaceae	2	4.08%	3	5.56%
Acanthaceae	2	4.08%	2	3.70%
Lamiaceae	2	4.08%	2	3.70%
Rhmnaceae	2	4.08%	2	3.70%
Other	22	44.90%	22	40.74%
Total	49	100.00	54	100.00%

List of medicinal plants used for Human ailments treatment in the study area of Assosa District were indicated in Appendix 1. Where the abbreviations indicated as: HBT=Habit (Hg- Home garden, Rs- Road side, W-Wild, Fl- Farm land). Language RU (Rutana), OR (Afaan Oromoo), AM (Amharic). HB=Habit (H- Herb, S- Shrub, Li- Liana, T-Tree, Ep- Epiphyte, ST- Shrub/small tree).

Regarding the habit of medicinal plants identified, shrubs took the highest proportion being represented with 20 species (37%), followed by 16 tree species (29%), 15 herbaceous species (26%). However, lianas and epiphytes took the least rank with three and one plant species, respectively (Figure 7).

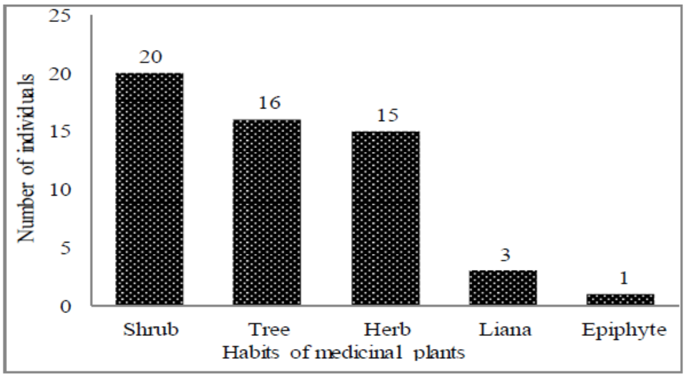

Figure 7. Habits of medicinal plants with their respective number of individuals

Parts of plants and their condition used for the preparation of traditional medicines

The most commonly used plant parts for herbal preparation in the present study area were also leaf (45%) for 35 preparations, followed by root 23% for 18 preparations, which shows that leaf was the most harvested plant part of many medicinal plants species for medicinal use. However, the second most harvested plant part was the root (Figure 8), which results in the whole removal of the mother trees, leading to the total destruction and extinction of the plants.

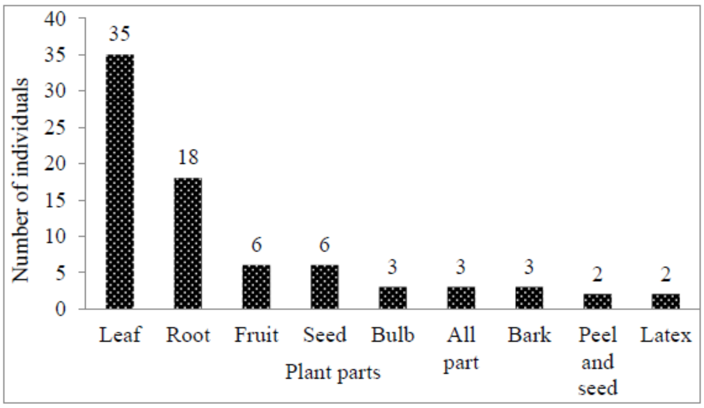

Figure 8 Plant parts used for traditional medicine preparation

Additionally, 64% medicinal plants were prepared from fresh plant materials, followed by 36% dry form. Concerning the habitats of medicinal plants, 21species (39%) were collected from wild while 17 species (31%) were from the home gardens.

Modes of preparation

The community of the study area employed various methods of preparation and application of traditional medicine for different types of diseases. The most commonly used method of remedy preparation was powdering (24%), followed by pounding (20%) (Figure 9).

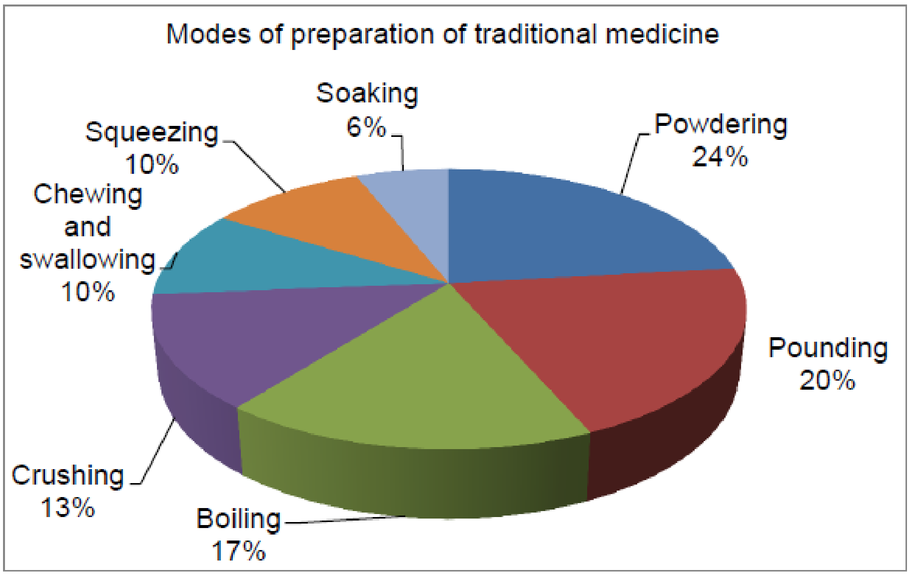

Figure 9 Different modes of preparation of traditional remedies from medicinal plants

Route of administration

A medication administration route is often classified by the location at which the drug is applied, such as oral, dermal, eye, nasal (sniffing), inhalation, and ear. In this study most commonly used route of administration was oral application for 42 preparations (52%), dermal application for 29 preparations (36%) nasal /Inhalation/ for seven preparations (9%) and optical application for two preparations (3%) respectively. As the result shows, oral administration was predominantly used in the study area. The methods of preparation and application vary based on the type and actual site of disease treated.

The herbal traditional medicines could also be prepared by using different solvents or additives like honey, water, and milk or prepared alone. However, in the present study area, 48% of the traditional medicines were prepared alone (without any additives); the other 23% and 12% of the medicines were prepared by using water and butter or Vaseline (Table 6).

Table 6. Solvents and additives used

Solvent or additive	Number of informants	Percentage (%)
Water	19	23.46%
Butter/Vaseline	10	12.35%
Honey	6	7.41%
Sugar	5	6.17%
Milk	2	2.47%
Alone (without mixing)	39	48.15%

Ranking of medicinal plants for their effectiveness

As reported by informants, medicinal plants were very effective in treating certain diseases. Even patients preferred to visit local healers than modern medicine. For instance, diseases such as cellulitis (locally called 'Lifie), rabies, spider poison, liver, Rh problem, hemorrhoids, mental illness, evil eye ('Buda'), and migraine were mostly cured by traditional treatments in the study area. Based on these general arguments, some of the plant species identified in the present study area were made to rate using some respondents of the local people using different ranking methods as mentioned before and discussed here after. Results of simple preference ranking for six medicinal plants against Hemorrhoid (6 stands for the most preferred medicinal plant, whereas 1 is for the least one) is indicated in Table 7.

Table 7. The result of simple preference ranking for six medicinal plants against Hemorrhoid

	R	E	S	P	O	N	D	E	N	T S
Medicinal plant species	R1	R2	R3	R4	R5	R6	R7	R8	Total	Rank
Clematis hirsuta Perro & Guill	3	1	3	5	6	5	6	4	33	1st
Physalis lagascae Roem. & Schult.	6	4	2	4	1	6	4	5	32	2nd
Senna baccarinii (Chiov.) Lock	2	3	4	5	6	3	5	1	29	3rd
Ferula communis L.	4	5	6	1	3	4	2	3	28	4th
Achyranthes aspera L.	5	6	1	3	4	3	1	1	24	5th
*Calotropis procera (*Ait.)Ait.f	1	2	5	2	5	2	1	2	20	6th

Direct matrix ranking

Based on the information gathered from the informants, six multipurpose tree species were selected out of the total medicinal plants and six use diversities of these plants were listed for 8 randomly selected informants to assign use values to each species. The six use values include medicinal, food, firewood, construction, charcoal, and furniture. Based on this, the most multipurpose medicinal plant was *Cordia africa* followed by *Eucalyptus citriodora,* whereas the least multi use medicinal plant was *Mangifera indica* (Table 8). On the other hand, from those six use values, fire wood followed by medicinal use are the most used value by the local community of the study area.

Table 8. Direct matrix ranking of medicinal plants with different uses *Key: CA= Cordia africana, CM= Croton macrostachyus, EC=Eucalyptus citriodora, MI=Mangifera indica, MA=Melia azendarach, MS=Moringa stenopetala*

Uses	*CA*	*CM*	*EC*	*MI*	*MA*	*MS*	Total	Rank
Medical use	3	5	4	3	4	4	23	2nd
Food	3	0	0	5	0	1	9	5th
Charcoal	4	3	4	2	3	4	20	4th
Construction	4	3	4	2	4	4	21	3rd
Furniture	5	3	5	1	4	3	21	3rd
Fire wood	4	5	5	3	5	2	24	1st
Total	23	19	22	16	20	18		
Rank	1st	4th	2nd	6th	3rd	5th		

Informant consensus factor (ICF)

This study has resulted in ICF values ranging from 0.50 to 0.96 (Table 9). The results showed that the ICD-10 category diseases of the eye and adnexa had the highest ICF value (0.96) or agreement by the informants among all the ailment categories, while the lowest ICF value (0.50) was obtained for injury, poisoning and certain other consequences of external cause's factors influencing health status and contact with health services disease category in the study area.

Table 9. Informant consensus factor (ICF)

Diseases category	nur	nt	ICF
Diseases of the eye and adnexa	29	2	0.96
Diseases of the digestive system	42	4	0.92
Diseases of the nervous system	24	3	0.91
Symptoms, signs and abnormal clinical and laboratory findings not elsewhere classified	40	5	0.89
Certain infectious and parasitic diseases	25	4	0.87
Diseases of the skin and subcutaneous	47	9	0.82
Disease of the respiratory system	34	7	0.81
Diseases of the systemic circulation*	34	8	0.78
Endocrine, nutritional and metabolic diseases	14	4	0.76
Diseases of the genitourinary system and ovulatory disorders	21	9	0.60
Injury, poisoning and certain other consequences of external causes**	23	12	0.50

*=Referring to the blood and blood-forming organs and certain disorders involving the immune mechanism;

**=Referring to factors influencing health status and contact with health services

Fidelity level (FL) of medicinal plants

The fidelity level of medicinal plants was calculated for the most frequently reported ailments to evaluate the species consensus. Those diseases reported frequently were the followings: hemorrhoid, evil eye, dermal/skin disease/wound, gastric ulcer, diabetes, and Cholesterol. In the present study, the highest FL of 100% was recorded for medicinal plant species such as *Clematis hirsuta* for healing hemorrhoid, and *Portulaca sp*. for healing gastric ulcer. The fidelity level was 97% for *Echinops kebericho* for healing evil eyes and *Abelmoschus esculentus* for healing Gastric ulcers (Table 10).

Table 10. Fidelity levels of medicinal plants

Diseases	Plant species	Ip	Iu	FI
Hemorrhoid/wart	*Achyranthes aspera* L.	4	5	80
	Calotropis procera (Ait.)Ait.f	3	5	60
	Clematis hirsuta Perro & Guill	2	2	100
	Ferula communis L.	1	2	50
Evil eye	*Echinops kebericho* Mesfln	35	36	97
	Securidaca longepedunculata Fresen.	21	22	95
Dermalskin diseases/wound	*Coffea arabica* L	32	35	91
	Croton macrostachyus Del	15	22	68
Gastric ulcer	*Portulaca sp.*	38	38	100
	Abelmoschus esculentus (L.) Moench	28	29	97
Diabetes	*Coriandrum sativum* L	5	7	71
	*Moringa stenopetala (*Baker f.) Cufod.	32	35	91
Cholesterol	*Citrus limon (*L.) Burn.f.	12	15	80
	Coriandrum sativum L.	5	7	71

Marketability of medicinal plants

There were reports of medicinal plants being sold in open markets solely for their medicinal use. However, some medicinal plants were indicated to be sold in local market also for their use as food, spices, and beverages. Hence, based on the observation *Abelmoschus esculentus* (food), *Allium sativum* (spice), *Carica papaya* (food), *Coriandrum sativum* (spice), *Rhamnus prinoides* (additive for fermented beverages), *Ruta chalepensis* (spice), *Securidaca longepedunculata* (medicinal), *Echinops kebericho* (medicinal) *and Zehneria* scabra (medicinal) were the major plant species sold on the markets of the study area*.* The practitioners around rural kebeles bring their medicinal plants on Saturday to the market to sell and get additional incomes for their families.

Threats and conservation of medicinal plants in the study area

In this study area, as reported by informants, human-related activities such as firewood collection, medicinal use, charcoal production, construction, over exploitation and urbanization were recorded as the main threats to the existence of medicinal plant species. In this respect, the plant species with multipurpose uses were highly affected. For instance, as observed during the field survey, medicinal plants such as *Cordia africana* and *Eucalyptus citriodora* are highly threatened by their being cut by local people from their natural forests deliberately to expand their agricultural and investment activities, secure their food and medicinal demands, and generate their additional incomes. They also prefer C*. africana* and *E. citriodora* fire wood, medicinal use, construction and charcoal production. Moreover, the results show that most of the practitioners of the study area depended on wild area to obtain their medicinal plants, which also increase the rate of threat on medicinal plant species. Furthermore, the result of the study shows that the root of the medicinal plant was used highly next to the leaf, leading to over exploitation of roots and the whole removal of the mother medicinal plants. Most of the medicinal plants visited during the market survey were also the roots (Figure 10).

Figure 10. Photo taken during the market survey and showing harvesting of root parts

Results of cluster analysis and ordination

Cluster analysis encompasses a number of different classification algorithms, which seek to organize a given data set into homogeneous subgroups (Woldu 2017). Hence, cluster Analysis is an exploratory data analysis tool, which aims at sorting different objects into groups in a way that the composition of the group varies most between groups and varies least within groups. It was noted that methods for clustering items (respondents or medicinal plant species) depend upon how similar (or dissimilar) the items are to each other. Similar items are treated as a homogeneous group, whereas dissimilar items form additional groups.

Much of the output of a cluster analysis is visual, with the results displayed as scatterplots, trees, silhouettes or dendrograms. When the objects within a group are very similar this can be described as internal cohesion (or homogeneity); when there is a large dissimilarity between groups this can be referred to as external separation (or isolation). A data set which when subjected to clustering results in high internal cohesion and high external separation can be considered to have distinct groups. Therefore, agglomerative hierarchical classification using reordered dendrogram for the 76 respondents and 54 medicinal plant species grouped the respondents into 10 clusters (groups) based on their medicinal plant knowledge and similarity of medicinal plant species scores (Figure 11).

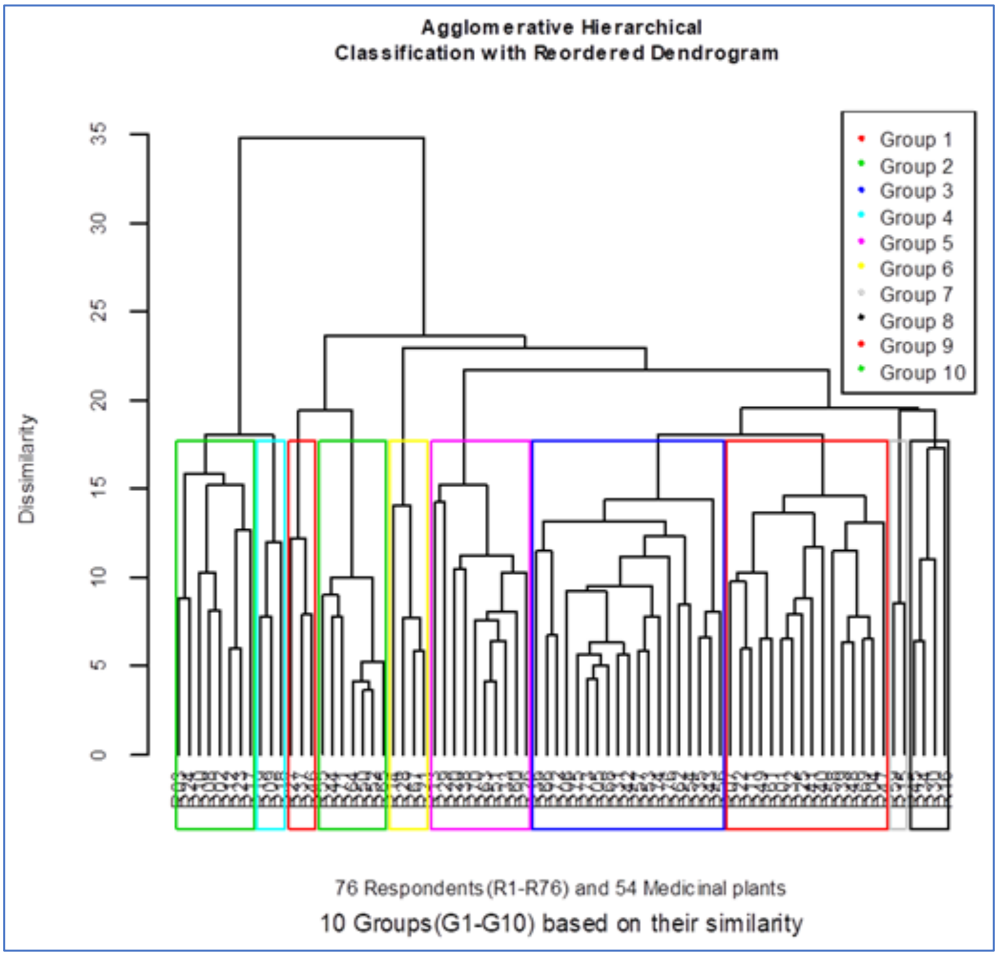

Figure 11. Agglomerative hierarchical classification using reordered dendrogram

Redundancy Analysis (RDA) ordination with sociodemographic variables where medicinal plant species scores scaled by eigenvalues revealed that sociodemographic variables, except education, showed strong correlation with informant category and their medicinal plant knowledge (Figure 12).

Similarly, RDA ordination for use value variables with species scores scaled by eigenvalues showed various degrees of significances of use value diversity variables (Figure 13)

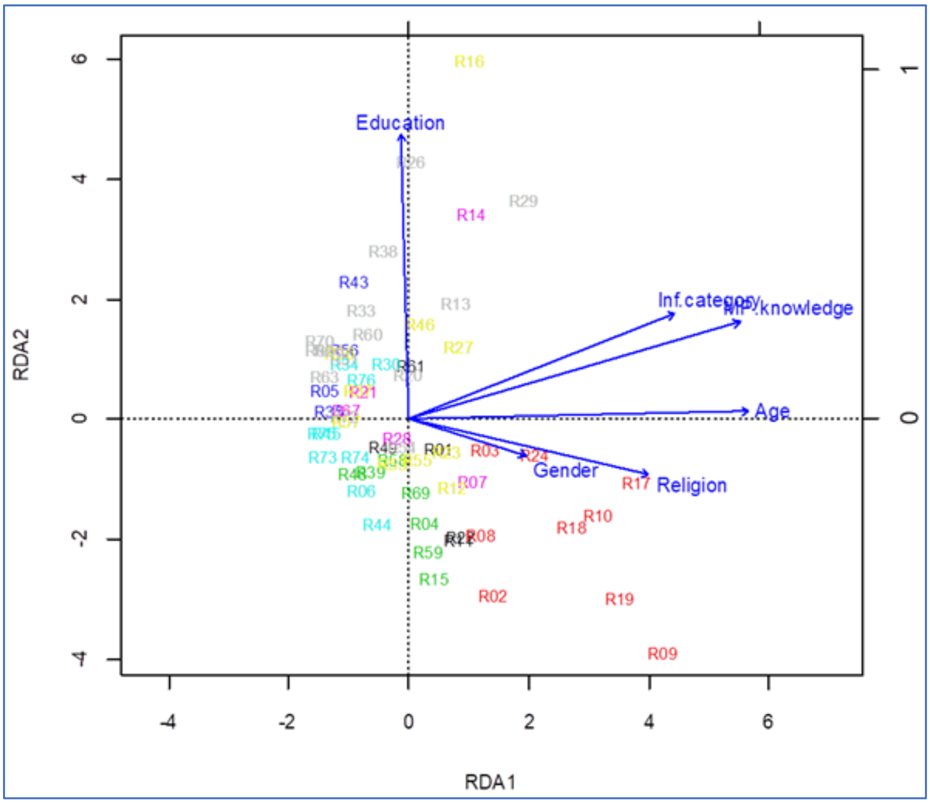

Figure 12. RDA with sociodemographic variables: species scores scaled by eigenvalues

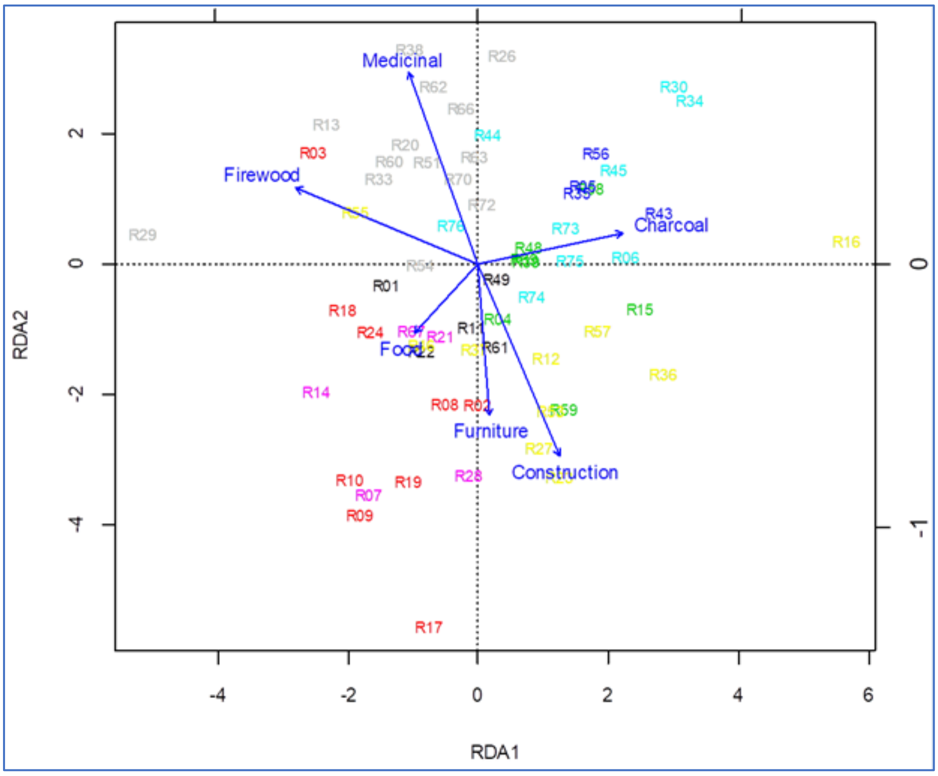

Figure 13. RDA for use value variables with species scores scaled by eigenvalues

The use of medicinal plants for fire wood has relatively the highest destructive potential (p=0.020) in this regard. Moreover, RDA ordination of informants' knowledge (diseases mention, multiuse mention other than medicinal use and medicinal use mention) with regard to sociodemographic variables (age, informant category, religion and gender) showed various degrees of relationships (Figure 14).

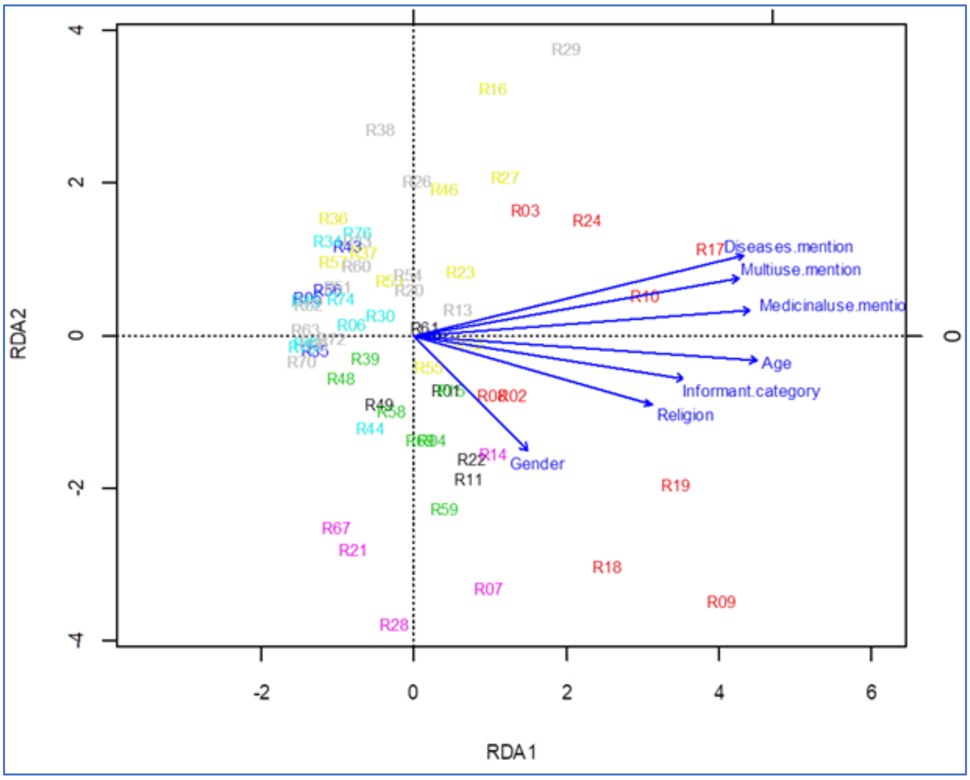

Figure 14. Ordination of Informants' knowledge with sociodemographic variables

Deterended Correspondece Analysis (DECORANA) was used to display both species and respondents biplot. Sociodemographic variables were fitted into ordination scatterplots to depict the plant human relationships (Figure 15). It showed that there was observed dissimilarities among respondents constrained by sociodemographic variables such as age, informant category, religion, gender and education.

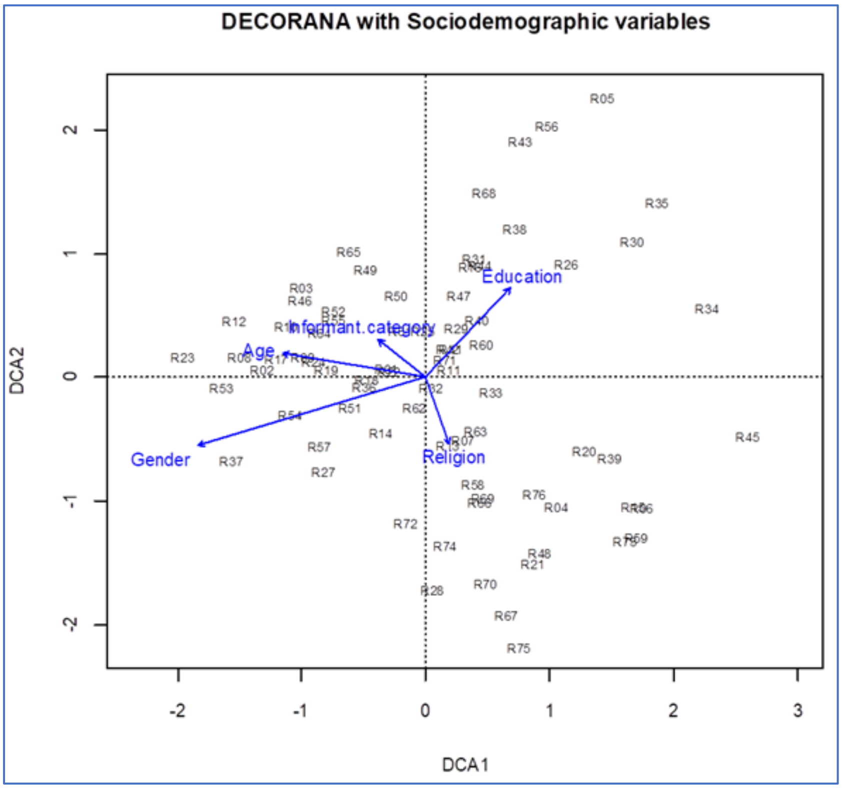

Figure 15. DECORANA with sociodemographic variables

Principal Coordinates Analysis (PCoA) is a method to explore and visualize similarities or dissimilarities of data. The level of similarity among causes in complex species-respondent dataset constrained by sociodemographic variables indicated that there was a 97.82% similarity among causes in the complex dataset (Figure 16). It showed goodness of fit, a statistical test that tries to determine whether a set of observed values match those expected values under the applicable model.

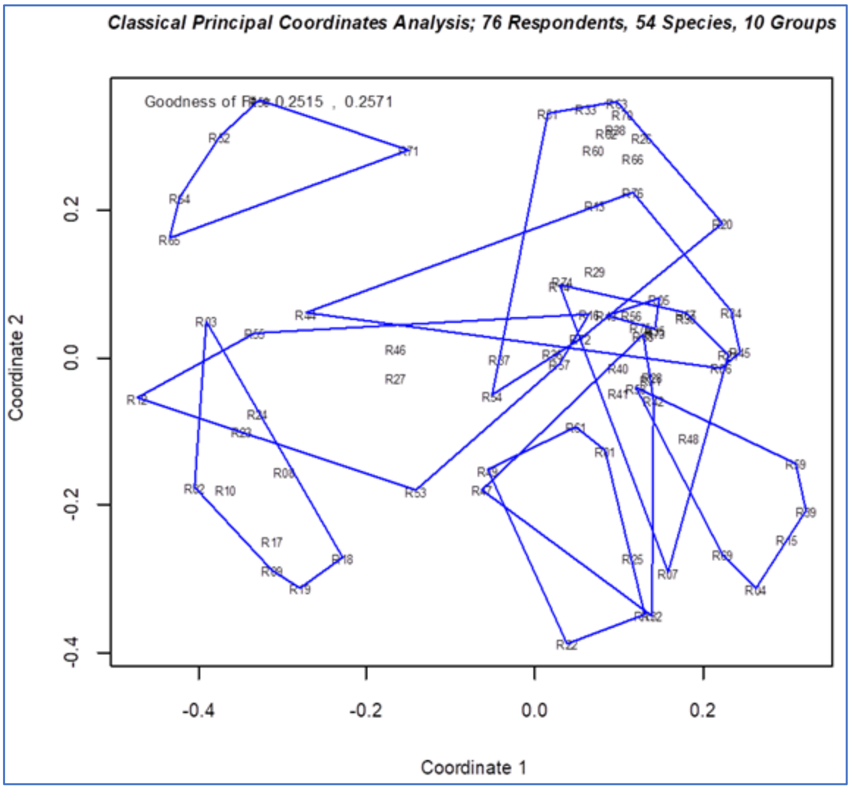

Figure 16. Classical Principal Coordinate Analysis (PCoA)

Results of field observations and informants' qualitative responses

Conservation is the protection, preservation, management, or restoration of wildlife and natural resources such as forests. In this study area, conservation of medicinal plants was weak. Most of the practitioners depended on forest to drive their medicinal plants and used them for charcoal production. However, some traditional practitioners had started to conserve medicinal plants by growing them in home gardens; for instance, *Ruta chalepensis*, *Abelmoschus esculentus*, *Citrus limon*, *Allium sativum*, and *Ocimum lamiifolium* were cultivated by those practitioners.

As also from field observation during the study period, the home gardens were fenced and the medicinal plants were protected from grazing and unwise harvesting. The practitioners of the city also reported as they need land to undertake conservation activities for the threatened medicinal plants and to introduce other medicinal plants growing in other areas so that they tried to submit their proposal to Assosa municipality, despite not yet getting positive responses.

To conclude, two contrasting viewpoints observed; in one hand, the conservation practice of the rural community is less; on the other hand, the practitioners found in the city have good activity of cultivating medicinal plants in their home gardens.

Discussion

Sociodemographic characteristics and use diversity variables

Ordination involves reduction of dimensionality to obtain simplicity for better understanding (Hoft & Lykke 1999). Use value indices were complemented with statistical tests, modeling and ordination methods to see the structure of relationships among plants and humans (Kikvidze & Bussmann 2024). Sociodemographic characteristics and different uses reports of respondents analyzed by different ordination methods showed various correspondence spaces between sociodemographic variables and different medicinal plant use reports. In the current study, variables such as age, informant category and religion showed higher significance than education and gender at $P \leq 0.05$ level of significance difference (Table 2). The ANOVA.CCA test for significance of use value variables also showed that the use of medicinal plants as source of fire wood was statistically higher (p=0.020) (Table 3). The difference between medicinal plant use and medicinal plant knowledge was also highly reflected in the current study as already stated in (Kikvidze & Bussmann 2024). Mouzazi *et al*. (2024) also reported that herbalists and practitioners of traditional healers have a low level of education and scientific knowledge with regard to dosage and parts used as compared to educated people.

Diversity and habits of the medicinal plants

The study identified 54 medicinal plants from 49 genera and 33 families in the area. In comparison, other regions reported varying numbers: 83 in northern Amhara (Getnet *et al.,* 2016), 32 in western Shewa (Getaneh & Girma, 2014), 44 in Somali Region (Wubu *et al*., 2023), and 188 in southern Ethiopia (Zemede *et al.,* 2024). Tamene *et al.* (2023) documented 189 species in southern Ethiopia. The lower diversity in most areas, except the south, is attributed to deforestation caused by urbanization, firewood collection, charcoal making, and overharvesting. Habitat loss from agricultural expansion, road construction, and investments also poses significant threats (Moges, 2019).

Adugna (2014) reported as the Regional State of Benshangul Gumz has diverse topography and climate. Yet, although – "*Kola*", "*Dega*", and "*Woyna-Dega*" agro climatic zones are there, "about 75% of the State is classified as "Kola" (law lands), which is below 1500 m.a.s.l. As a result, the majority of the medicinal plants (20 species, 37%) collected from the study area belonged to shrub species, followed by 16 tree species (29%). This finding is similar to the finding of Wubu *et al* (2023) from Somali Region, which might be due to their similarity in their agro climatic zones. However, the finding is contrary to the general pattern seen in most medicinal inventories carried out in Ethiopia (Amenu 2007, Getaneh & Girma 2014, Morka & Duressa 2021), where herbaceous medicinal plants are dominant. This might be because that the agro climatic condition of Benshangul Gumuz Region, especially Assossa district is low land area, where shrubs and trees grow more widely than the herbaceous ones. This may be explained by the fact that shrubs are perennial in the arid or sub-arid environments and may be available for use as medicinal plants.

Parts of plants and their condition used for the preparation of traditional medicines

Medicinal properties derived from plants parts plant including leaves, roots, bark, fruit, seeds, flowers (https://www.fs.usda.gov/wildflowers/ethnobotany/medicinal). The different parts of plants can contain different active ingredients within one plant. Thus, one part of the plant could be toxic, while another portion of the same plant could be harmless. Their dominant use could also be attributed to the ease with which they are harvested, whereby they have positive sides as they are re-vegetated compared to the root parts of the plants, despite affecting the plant growth due to their being photosynthetic organs in plants. These results are in agreement with other similar studies conducted elsewhere in Ethiopia (Amenu 2007, Enyew *et al*. 2014, Kassa 2009, Moges 2019) and in Kenya (Maiyo *et al*. 2024), where leaves were the most frequently used.

Additionally, 64% medicinal plants were prepared from fresh plant materials, followed by 36% dry form. This finding is also in line with the finding of Amsalu *et al*. (2018) and Enyew *et al.* (2014), who reported that the majority of the remedy preparations was in fresh form, which might be due to higher ingredient contents of in fresh form than the dry one, which in turn, could enable the plants to be effective in preventing the respective health problems.

Habitats of medicinal plants

This study highlights that a significant number of local practitioners primarily rely on the wild for sourcing medicinal plants, with home gardens serving as a secondary option. Specifically, most practitioners residing in the rural kebeles of Assossa heavily depend on the natural environment for obtaining these plants. Similarly, Awas (2007) reported that 70% of medicinal plants in Benshangul Gumuz were collected from the wild. The current findings also reveal that indigenous people in the Assossa district rely predominantly on the wild for medicinal plants. Conversely, practitioners in urban areas have adopted better practices of cultivating medicinal plants in their home gardens. This pattern suggests that forests in the rural kebeles of Assossa District are being overexploited by traditional practitioners for medicinal purposes. These findings align with other ethnobotanical studies conducted in Ethiopia (Alemayehu *et al*., 2015; Kassa *et al*., 2020), which also identified wild habitats as the primary source of medicinal plants.

Modes of preparation

The community of the study area employed various methods of preparation and application of traditional medicine for different types of diseases. The preparations vary based on the type and actual site of disease treated. The most commonly used method of remedy preparation was powdering (24%), followed by pounding (20%), This finding is contrary to the general pattern seen in most medicinal inventories (Megersa *et al.* 2013, Tamene et *al.* 2020, Jima & Megersa 2018), where crushing was the most common method of herbal remedial preparation. This also shows that there are various types of commonly used methods for preparing herbal medicines from medicinal plant species in different parts of Ethiopia.

Route of administration

A medication administration route is typically categorized based on the location where the drug is applied, such as oral, dermal, nasal, eye drops and ear medication. The selection of an administration route depends not only on convenience but also on the drug's properties and pharmacokinetics (Kim & Jesus, 2023). In this study, the oral route of administration was the most commonly employed. This preference was primarily due to the nature of the disease, the characteristics of the medicinal plants, and the effectiveness of the treatment. These findings align with those of other ethnobotanical studies conducted in different regions of Ethiopia (Alebie & Mehamed, 2016; Wubu *et al*., 2023), which also identified oral administration as the most frequently used route. Furthermore, the methods of preparation and application varied depending on the type of disease and its specific site of treatment.

In the present study, 48% of the traditional medicines were prepared alone (without any additives); the other 23% and 12% of the medicines were prepared by using water and butter or Vaseline. This implies that although the practitioners reported that those additives had their own values in increasing the effectiveness of the herbal traditional medicines, the plants growing in very lowland areas are supposed to be highly concentrated in their chemical components so that they might not demand mixing them with other plant species.

Lack of precise dosage is one of the challenges that the traditional medicinal plants cannot be having reliability among some communities. This is because of weak integration between the modern and traditional medicine. Of course, most of the practitioners of the present study area used different methods to measure the dosage of the medicine including cups, spoons, glass, and other measurement techniques. Similar measurement units were reported by Teklay *et al.* (2013). However, the dosage given was variable among age and longevity of the disease in the study area. These were done mostly for orally admitted medicine. On the other hand, for dermally applied medicines, they were not mostly using dosage measurement, as they reported that the side effect of herbal medicine on dermal is less. Generally, in this study area, the reliability of the local community on dosage application of remedies is less. Almost all authors agreed with this finding (Kassa *et al*. 2020, Jima & Megersa 2018, Tamene *et al*. 2020). Hence, the dialog in dosage variability of remedies for the same disease among practitioners and local community continued (Teklay *et al*. 2013).

Additives in pharmaceuticals serve numerous functions. Drug additives commonly serve as agents of coloring, flavoring, emulsification, thickening, binding, palatable and preservation (Vrancheva *et al*. 2018). The herbal traditional medicines could also be prepared by using different solvents or additives like honey, water, and milk or prepared alone. Such application of additives was also reported by other authors (Getaneh & Girma 2014; Getnet *et al.* 2016). The use of different solvents or additives in herbal remedies preparations as indicated by the current study implies that although the practitioners reported that those additives had their own values in increasing the effectiveness of the herbal traditional medicines, the plants growing in very lowland areas are supposed to be highly concentrated in their chemical components so that they might not demand mixing them with other plant species for dilution purposes. The reason behind plants growing in lowland areas are highly concentrated, as reported by the respondents, needs further phytochemical studies on medicinal plants in relation to different climatic factors and agro-ecological conditions for possible scientific proof.

Ranking of medicinal plants for their effectiveness

Regarding the effectiveness of medicinal plants, several studies have indicated that certain illnesses are believed to be either untreatable by modern healthcare or better managed through traditional remedies using medicinal plants. According to local informants, medicinal plants were highly effective in treating specific diseases. In fact, many patients preferred seeking treatment from local healers rather than relying on modern medicine. Based on these observations, some plant species identified in the study were evaluated through ranking methods involving local respondents, as previously described and discussed further in this study.

Ranking and scoring of medicinal plants were employed to compare different medicinal plants that are preferred by local people than the others based on their effectiveness in treating their diseases. In this study, the discussions were, therefore, made with two groups of 16 participants, (8 respondents for preference and the other 8 participants for direct matrix ranking) for who were drawn from each selected kebeles (about two individuals per kebele) by selecting the six medicinal plants used to treat hemorrhoid *Clematis hirsuta* Perro & Guill were the most preferred medicinal plant to treat hemorrhoid disease, whereas the least preferred medicinal plant was *Calotropis procera* (Ait.)Ait.f

Similar studies confirmed that the use of some medicinal plants to treat health problems was frequently reported while the use of others was less reported (Megersa 2023). For instance, a review by Fekadu *et al*. (2023) on the potential of Ethiopian medicinal plants to treat emergent viral diseases revealed that *Zingiber officinale, Allium sativum, Croton macrostachyus, Ocimum lamiifolium* and *Ruta chalepensis* are among the most cited medicinal plants. On the other hand, *Clerodendrum myricoides*, *Cordia africana, Euphorbia abyssinica, Foeninculum vulgare*, *Momordica foetida* and *Nicotiana tabacum* were among the leas cited medicinal plants to treat viral diseases. A systematic review on traditional medicinal plants used in treatment of tuberculosis in Ethiopia also revealed that the most frequently reported plant species belong to family Lamiaceae, Euphorbiaceae, Cucurbitaceae and Fabaceae (Getachew *et al.* 2022). The authors also reported that *Croton macrostachyus, Allium sativum* and *Myrsine african* were the most often mentioned anti-TB medicinal plants. Hence, the current study is in line with previous studies.

Direct matrix ranking

Based on the information gathered from the informants, six multipurpose tree species were selected out of the total medicinal plants and six use diversities of these plants were listed for 8 randomly selected informants to assign use values to each species. The six use values include medicinal, food, firewood, construction, charcoal, and furniture. Based on this, the most multipurpose medicinal plant was *Cordia africa* followed by *Eucalyptus citriodora,* whereas the least multi use medicinal plant was *Mangifera indica*). On the other hand, from those six use values, fire wood followed by medicinal use are the most used value by the local community of the study area. Studies conducted in different parts of Ethiopia showed that medicinal plants have multipurpose values where in the meantime subjected to treats arising from the multipurpose needs (Megersa *et al*. 2013, Yimam *et al*. 2022). For instance, the authors reported that fire wood collection, charcoal making and the use of plants for construction purposes were among the major treats to the medicinal plants.

Informant consensus factor (ICF)

The informant's consensus factor was also employed to look into the level of homogeneity among informants for the plants to be used for each ailment category (Alexaides 1996). The current study indicated that diseases categories such as Diseases of the eye and adnexa, Diseases of the digestive system and Diseases of the nervous system are among the top three health problems with ICF score of greater than ninety percent whereas genitourinary system and ovulatory Disorders; injury, poisoning and certain other consequences of external causes have the lowest ICF scores of less than ten percent (Table 7). It indicates that diseases with the highest informant consensus factor are the most prevalent in the study area and vice versa. The current finding supports similar studies by Megersa *et al*. (2023, 2013) and Yimam *et al.* (2022).

Fidelity level (FL) of medicinal plants

The fidelity level of medicinal plants represents the relative healing potential of medicinal plants against a given ailment. In the current study, the fidelity level of medicinal plants was calculated for the most frequently reported ailments to evaluate the species consensus. Those diseases reported frequently were the followings: hemorrhoid, evil eye, dermal/skin disease/wound, gastric ulcer, diabetes, and Cholesterol. Moges (2022, 2019) also reviewed or reported many of these diseases affecting Ethiopian people. Accordingly, in the present study, the highest FL of 100% was recorded for medicinal plant species such as Clematis hirsuta for healing hemorrhoid, and Portulaca sp. for healing gastric ulcer. The fidelity level was 97% for *Echinops kebericho* for healing evil eyes and *Abelmoschus esculentus* for healing Gastric ulcers. These all imply that those species are best for treating their perspective diseases so that they should be protected and widely propagated in home gardens and farmlands, besides conserving them in their in-situ.

Marketability of medicinal plants

Market is a place where medicinal plants, fruits and other plant products are sold. Thus, markets are rich sources of ethnobotanical information. According to Martin (1995) the economic study of biological resources cannot be complete without a detailed survey of the plants and animals sold in local market places. Many medicinal plants, ornamentals, wild foods, edible insects and other products have a strictly regional value which can only be discovered by talking with producers, sellers and consumers.

There were reports of medicinal plants being sold in open markets solely for their medicinal use. However, some medicinal plants were indicated to be sold in local market also for their use as food, spices, and beverages. Hence, based on the observation *Abelmoschus esculentus* (*food*), *Allium sativum* (spice), *Carica papaya* (food), *Coriandrum sativum* (spice), *Rhamnus prinoides* (additive for fermented beverages), *Ruta chalepensis* (spice), *Securidaca longepedunculata* (medicinal), *Echinops kebericho* (medicinal) *and Zehneria scabra* (medicinal) were the major plant species sold on the markets of the study area. The practitioners around rural kebeles bring the medicinal plants to markets to sell and get additional incomes for their families.

Threats and conservation of medicinal plants in the study area
Human-related activities such as fire wood collection, medicinal use, charcoal production, construction, over exploitation and urbanization were recorded as the main threats to the existence of medicinal plant species. In this respect, the plant species with multipurpose uses were highly affected. For instance, it was observed that medicinal plants such as *Cordia africana* and *Eucalyptus citriodora* are highly threatened by their being cut by local people from their natural forests deliberately to expand their agricultural and investment activities, secure their food and medicinal demands, and generate their additional incomes. They also prefer *Cordia africana* and *Eucalyptus citriodora* for fire wood, medicinal use, construction and charcoal production. Moreover, most of the practitioners of the study area depended on wild habitats to obtain their medicinal plants, which also increase the rate of threat on medicinal plant species. Furthermore, the root of the medicinal plant was used highly next to the leaf, leading to over exploitation of roots and the whole removal of the mother medicinal plants. Most of the medicinal plants visited during the market survey were also the roots. The findings of the current study supported similar findings by previous studies (Megersa *et al*. 2013, Yimam *et al*. 2022).

Ordination methods to display the structure of relationships among plants and humans
Ordination is a collection of techniques which transform vector observations into a new set of vector observations (Woldu 2017). Ordination analysis did not allow visualization of qualitative differences (Dip & Vattuone 2024). Hence, numerical values of medicinal plant use reports by each respondent and the rated destructive potential of medicinal plant use diversity variables (multiple plant uses in addition to its medicinal use) were used to construct dendrogram and ordination graphs. Knowledge differences can be observed as one goes higher the dendrogram in the y-axis with increasing dissimilarity. The grouping was further constrained by sociodemographic variables (age, gender, informant category, education and religion) which latter explained by ordination scatter points.

Pearson correlations coefficients showed the presence of significant associations among the informants at various levels based on sociodemographic factors. According to Zamin *et al.* (2024), moderate correlation was observed between respondent's age and traditional knowledge with non-significance and age is better predictor of indigenous knowledge than qualification supporting the current study.

As indicated in the ordination graphs, the more closely the arrows the more similar effects of sociodemographic variables. Moreover, arrow lengths show the degree of influences of the sociodemographic variable where the longest arrow is the most influential. It was supplementd by the value of Akaike Information Criterion (AIC) where the variable with the lowest AIC is the most influential. Hence, age is the most influential sociodemographic variable in the current case. The distances between arrows and their lengths supplemented by AIC values best explain the influences of sociodemographic variables on their ethno medicinal and ethnobotanical knowledge (diseases mention, multiuse mention and medicinal use mention) which is also among the dimensions of looking at the structure of relationships among plants and humans.

When correspondence analysis suffers from the arc effect of unimodal species curves in relation to informant responses, Deterended Correspondence Analysis (DECORANA) is appropriate to eliminate the problem (Borcard & Legendre). Hill & Gauch (1982) developed an improved ordination technique, DECORANA to eliminate the arc effect of Principal Coordinate Analysis (PCoA) due to unimodal species response curves.

The Meta Multidimensional Scaling (metaMDS) to display respondents and medicinal plant species was also used to find a goodness of fit measure statistic for observations (points) (https://rdrr.io/rforge/vegan/man/goodness.metaMDS.html). A plot of ordination distances versus observed dissimilarity showed that there was 73.70% goodness of fit (Figure 17). Hence, there was remarkable dissimilarity among respondents with regard to their responses as constrained by sociodemographic variables.

In summary, it was claimed that most ethnobotanical studies often ignored the incorporation of numerical analysis and ordination methods in their work. However, ordination methods are good representations of the structure of relationships among plants and humans (Kikvidze & Bussmann 2024). They show significant associations among medicinal plant knowledge and sociodemographic factors where heterogeneous sociodemographic profiles reflected (Ghobbour *et al*. 2024, Gum *et al.* 2024). In the meantime, such statistical rigor will help scientific validation of some of the plants that are used to manage health problems as alternative medicines and hence design biodiversity conservation priorities (Anas *et al*. 2024, Anbessa *et* *al.* 2024, Beyene *et al*. 2024, Gobvu *et al*. 2024). It was reported that implementation of measures to prevent cultural erosion, loss of traditional knowledge, conservation and protection of rare species from being overused thereby subjected to extinction need valid research based scientific information to design conservation, restoration and sustainable use of plant resources (Amjad *et al*. 2020, Dip *et al*. 2024, Gobvu *et al*. 2024).

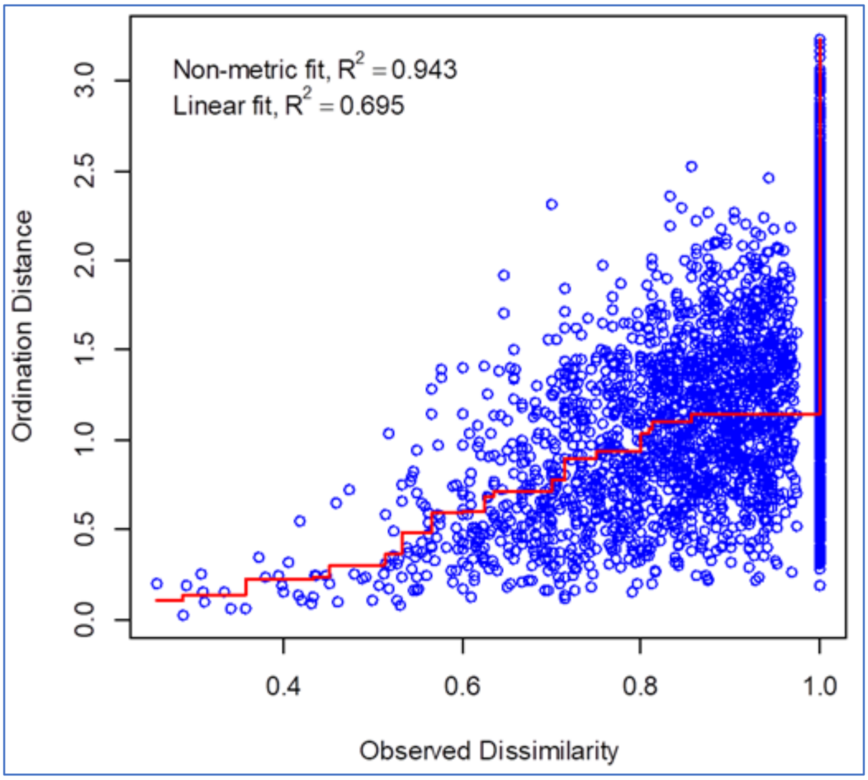

Figure 17. metaMDS to find goodness of fit measure for each informant

Other traditional healing methods used by the practitioners; Wagemt (cupping therapy)
During conducting this study, other traditional method of healing was assessed besides using the plant. This method is cupping therapy, which is locally called ''Wagemt'' in Amharic. This method is used to reduce different internal and external diseases by extracting the blood of human which is toxic in the affected area or which causes disease. Cupping is not randomly put on the skin of the patient rather depending on the treated ailment and the condition of the patient, as reported by key informants. As they also reported that the area of application is different for different diseases. According to the local healer, the cups were left in place anywhere from 10 to 30 minutes depending on the nature of the individual's condition.

Respondents say that they preferred to use plastic cups because they do not break as easily as pottery or deteriorate like bamboo, and they allow the local healer (the therapists) to see the skin and evaluate the effects of treatment. Based on the information from one of the key informants the method is very crucial to cure back pain, nerve problems, skin problems such as eczema and acne (locally called 'Chefie' and 'Bigure' in Amharic), high blood pressure, blood disorders, migraine. The method can also improve blood and lymph circulation, the immunity system of human body, muscle relaxation and overall health by removing the energy blockage and decreases or dispel internal toxins. In the study area wet cupping therapy, i.e., small skin incision and suction of blood using the cups, was widely used. After that, the practitioner gives immunity booster medicine prepared from plant material for the patient (Figure 18).

Conclusion
The study emphasized the significance of traditional medicine in primary healthcare systems. Key factors driving reliance on traditional medicine include a shortage of healthcare workers, the remoteness of healthcare facilities, and the high cost of modern healthcare services. Additionally, respondents reported that medicinal plants from lowland areas tend to have higher concentrations of active compounds. This observation calls for further phytochemical research to explore the impact of climatic and agro-ecological factors on the medicinal properties of these plants, providing a potential scientific basis for their use.

Some of the informants were not volunteers to easily share their knowledge. Therefore, special training for the practitioners must be given to make them conserve medicinal plants in their natural habitats (*in-situ*), and in their home gardens and farmland (*ex-situ*) for sustainable use of the resources. The governmental and non-governmental organizations should work in collaboration with those practitioners for supporting them in getting finance, training on their limitations, recognition and intellectual property rights by Ethiopian Intellectual Property Agency for continuity of the Indigenous Knowledge. Many of the urban dwellers were not using traditional remedies because of unsecured dosage, which indicates the weak integration between traditional healers and modern medicine experts. Hence, it is recommended to minimize the gap between the two by creating a strong integration between traditional healers and modern health professionals. This could also support to carry out scientific and pharmacological analyses on medicinal plants, which in turn give confidence to use the remedies with precise dosage.

Figure 18. Photo taken during wet cupping therapy undergo for the patient

Declarations

List of abbreviations_ AIC- Akaike Information Criterion**;** ANOVA.CCA-Analysis of Variance.Cannonical Correspondence Analysis**;** CCA**-** Canonical Correspondence Analysis**;** CSA-Central Statistical Agency of Ethiopia**;** DECORANA=Deterended Correspondence Analysis; FGD-Focus Group Discussion**;** FL-Fidelity Level**;** ICD-International Classification of Diseases; ICF-Informant Consensus Factor; MetaMDS-Meta Multidimentional Scalling; PCoA- Principal Coordinate Analysis; RDA-Redundancy Analysis; UNICEF-United Nations International Children's Emergency Fund; USDA-Unite State Department of Agriculture; WHO-World Health Organization

Ethics approval and consent to participate: A letter of cooperation for conducting the research on the behalf of the researchers was obtained from the Department of Biology, Debre Birhan University to Assosa District Administration Office. The local authorities considered the permission letter and acknowledged to conduct the research on medicinal plants in Assosa District.

Consent for publication: All people shown in images gave their consent to have the images published.

Availability of data and materials: All the data are presented in figures, tables and appendix in the manuscript. The data are also available with the corresponding authors.

Competing interests: The authors declare that they have no competing interests.

Funding: Debre Birhan University is highly appreciated for the financial support to conduct the research.

Authors' contribution: Yeron Zelalem selected the research title, conceptualized, designed the research; collected, analyzed, presented the data and prepared the manuscript. Admasu Moges and Zewdie Kassa helped in conceptualization, research design, data collection, manuscript preparation and proof reading. Zewdie Kassa also authenticated plant species; recategorized and analyzed the data in R Statistical Software using ordination methods, statistical tests and models.

Acknowledgments

Debre Birhan University is acknowledged for financial support to the research. The local communities, administrative bodies and traditional healers of Assosa District are also greatly appreciated for their unreserved support and collaboration during field data collection.

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Supplementary material- Checklist of semi structured-interview/questionnaires used as basis for discussion and interview

Dear respondent,

The objective of this semi-structured interview/questionnaire/group discussion is to collect ethnobotanical data that help to document the traditional use of medicinal plants used by indigenous people and associated indigenous knowledge to treat human health problems in Assosa District, Benishangul Gumuz Regional State, Ethiopia. Your kind response and cooperation is vital for conducting the research and is highly valued.

Date -----------------------

I. Informant detail

Name/ID/…………… Kebele …………………………....

Age …………….. Religion………………………..

Occupation……………………….. Marital status --------------

Sex…………………….. For how long have you lived in the area? ----------------

Educational status…………….. -------------------------------------

II. Ethnobotanical information from key informants

1. What are the main human health problems attaching the local people in your Keble?

____

____

2. Mention plant types used to treat a given disease in the area /give local names/.
   A) Local name of the plant --------------------------------
   B) Habitat of the plant ---------------------------------------
   C) Habit of the plant ---
   D) Type of disease treated ---------------------------------
   E) How is it used (dried or fresh)
   F) Part of the plant used -----------------------------------
   G) Mode of preparation-------------------------------------
   H) Dosage (Amount used) ----------------------------------
   I) For how long --------------------------------------
   J) Way of administered -----------------------A) Oral B) Nasal C) Inhalation
   K) Plant collected from -------------------- A) wild B) Home Garden C) Market
   L) Plant collection time -------------------------------------
   M) Conservation practices, and status ----------------------
   N) Side effect of the medicine and its antidote -----------------------------
3. Means of knowledge transfer (oral or document), and to whom______________________________
4. What other uses apart from the medicinal use do the medicinal plants have? Please circle all that apply:
   a/ Firewood b/food c/furniture d/construction e/ charcoal
5. What do you think are the major threats to medicinal plants in your area? Please circle all that apply:
   a/ the use of plants for various purposes as in indicated under #4 above b/habitat modification: urban expansion, agricultural expansion
   c/ Deforestation
   d/ others (please specify)
6. How do you perceive the present day availability of medicinal plants as compared to past two or more decades? Why?
7. Which medicinal plant in your area is the most preferred? Why?
8. List the most common human health problems in your area and associated medicinal plants used to treat them.

Human Health problems	Medicinal plants used to treat the health problems

III. Questions for focus group discussion for all informants

1. What types of disease are common in this area?
2. How do you preventing them?
3. The current status of medicinal plants in this area?

4. What is the gap between modern and traditional medicine in this area?
5. Is it governmental institution or organization work with traditional practitioners?
6. What are the threats on medicinal plants currently?
7. What are the traditional practices used to conserve the medicinal plants?
8. What are other traditional healing methods used in the community?
9. What are the ways of transferring the local knowledge among the community?
10. The cost of medicinal plants.
    11. What other uses apart from the medicinal use do the medicinal plants have? Please circle all that apply:
        a/ Firewood b/food c/furniture d/construction e/ charcoal
    12. What do you think are the major threats to medicinal plants in your area? Please circle all that apply:
        a/ the use of plants for various purposes as in indicated under #4 above b/habitat modification: urban expansion, agricultural expansion
        c/ Deforestation
        d/ others (please specify)
    13. How do you rate the treats to medicinal plants? Please rate each you specifies under Q#12 as: A/Very high B/ High c/ medium d/ low e/ very low
    14. How do you perceive the present day availability of medicinal plants as compared to past two or more decades? Why?
    15. Why do you think people prefer the use of medicinal plants instead of modern medicine to cure health problems in your area?

Thank you!