JOURNAL OF PLANT RESOURCES

Volume 23 Number 1

Government of Nepal
Ministry of Forests and Environment
Department of Plant Resources
Thapathali, Kathmandu, Nepal
2025

ISSN 1995 - 8579

Journal of Plant Resources, Vol. 23, No. 1

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Date of Publication: 2025 June

Cover Photo: **From top to clock wise direction.**

Acmella calva (DC.) R.K.Jansen (PC: Nishanta Shrestha)
Flower of *Clematis phlebantha* L.H.J.Williams (PC: Amrit K. C.)
Effect of DCM extracts of Leaf of *Parthenium hysterophorus* L. in wheat seedlings at concentration (PC: Chandra Bahadur Thapa)
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Inhibition zones of ethylacetate extract of *Tagetes minuta* L. against *Staphylococus epidermidis* (PC: Sujata Adhikari)

Citation:

Name of the author. (Year of publication). Title of the paper, *Journal of Plant Resources, 23* (1), pages.

ISSN: 1995-8579

Published By:

Publicity and Documentation Section
Department of Plant Resources (DPR), Thapathali, Kathmandu, Nepal.

The issue can be retrieved from
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Editorial

It is our pleasure to bring out the current issue of Journal of Plant Resources, Volume 23, Number 1, Year 2025, a continuation of research publication by the Department of Plant Resources. Nine peer reviewed articles based on original research, two review articles and one short communication have been incorporated in this issue. The articles have been categorized as ecology, ethnobotany and phytochemistry.

This issue intends to cover the research activities of the department as well as of other research organizations. We encourage the young researchers to pursue quality research and contribute to build scientific knowledge on plant resources. We would like to establish a link between the inference of scientific research and societies through dissemination of knowledge and information. We believe that the research findings will be useful to the scientific community as well as general public to update the information on recent activities& development of plant science in Nepal.

We would like to thank all peer reviewers whose critical comments and suggestions has helped to improve the quality of the journal. We would like to acknowledge the contribution of the contributors for their interest in publishing their valued work in this journal and looking forward for further cooperation and collaboration with this department.

We would like to apologize in advance for any mistakes in this issue and at the same time promise to improve the future issues based on your valued input.

Journal of Plant Resources (2025) **23**(1), 1-6
https://doi.org/10.3126/bdpr.v23i1.83322

Comparative Study of Fatty Acids Profiles in *Mangifera indica* L. Seed Butter Extracts from Different Regions in Nepal Using GC-MS

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Abstract

Mango is widely consumed for its pulp; however, the kernels are often discarded as a waste product despite their potential application. This research aims to study the potential application of the kernel in the production of butter. The study involves chemical profiling of butter extracted from kernels of mango collected from various regions of Nepal for their possible application in the confectionery industry as a cocoa butter replacement. Samples were collected from different districts, including Bardiya, Doti, Kathmandu, Tanahun, and Dhangadhi. The collected samples were cleaned, dried, and ground into powder. Butter was extracted employing the Soxhlet extraction technique using n-hexane as a solvent. The extracted mango seed butter was then analyzed using Gas Chromatography- Mass Spectrometry (GC-MS) to determine the fatty acid composition. Despite species-specific variations in concentration, octadecanoic acid (stearic acid), a crucial component of mango seed butter, was the most consistently found chemical in the samples studied. Some other important fatty acids such as oleic acid and palmitic acid were also present in the kernel of mango.

Keywords: Cocoa butter, Fatty acids, Mango kernel, Octadecanoic acid

Introduction

Mango (*Mangifera indica* L.) has been commercialized globally in more than 90 nations. It is commonly grown in tropical and subtropical regions of the world (Akhter et al., 2016). The growing global demand for mangoes and their processed products focuses on pulp for making juices, jams, and other items (Tharanathan et al., 2006). Consequently, mango seeds, a major by-product along with peels, account for 60% of the fruit and are discarded as waste. Despite this, mango seeds, containing 7.3-15% fat, can produce over 123,000 metric tons of oil (Abdalla et al., 2007). Mango seed contains a type of fat called mango kernel fat (MKF) (Solís-Fuentes & Durán-de-Bazúa, 2020). Mango kernel fat is a rich source of stearic and oleic acid (Wu et al., 2015). These fats are notable for their distinct physical and chemical properties, which make them highly suitable for manufacturing confectionery products (Naeem et al., 2019). Consequently, mango seed lipids have gained attention for their potential as a cocoa butter replacement. Where, four major fatty acids that include palmitic acid (24.4 %), stearic acid (33.6 %), oleic acid (37.0 %), and linoleic acid (C18:2, 3.4 %) account for more than 98 % of the total fatty acids in commercial cocoa butter (Lipp et al., 2001). Recently, studies have displayed the potential of mango as an alternative to cocoa butter with wide applications like pharmaceuticals, cosmetics, etc. due to its healthy fatty acids, antioxidants, and antimicrobial activity. Studies have shown that the main fatty acids found in mango kernel oil are about 45 % oleic acid and 38 % stearic acid (Wu et al., 2015). Oleic acid is an 18-carbon monounsaturated fatty acid, essential in human nutrition, and helps reduce triglycerides, LDL-cholesterol, total cholesterol, and glycemic index (Kittiphoom & Sutasinee, 2013). Also, the increase in stability over the oxidation of vegetable oil is attributed to oleic acid (Anwar et al., 2007). In addition to that, mango kernel seed extract also enhanced the oxidative stability of fresh-type cheese and ghee and extended their shelf life (Melo et al., 2019).

Cocoa butter is a light yellow fat that is obtained from the cocoa bean of the cocoa plant (*Theobroma cacao* L.). It is highly demanded by the food, pharmaceutical, and cosmetic industries. Moreover, it is unique among vegetable fats due to its composition and crystallization behavior. Cocoa butter contains 33.5% oleic acid, 25% palmitic acid, and 33% stearic acid, which accounts for above 80% of the total fatty acids in commercial cocoa butter (Chen et al., 1989; Shekarchizadeh et al., 2009). However, the challenges in cultivation, limited availability, and high demand contributed to the expensive nature of cocoa butter (Darmawan & Mutalib, 2024).

While the chemical composition of mango seed butter has been explored to some extent, there is a noticeable gap how this composition varies across different locations. The primary objective of this research is the extraction of the butter from the mango kernel sourced from various geographical regions and its analysis for fatty acid composition, to understand how the plant origin affects the composition of kernel fat. The study also aims to compare the composition of cocoa butter with that of mango kernel butter to explore its potential as a viable alternative. The findings indicate significant properties that highlight its suitability as an alternative to cocoa butter and promising application in the confectionery industry. Ultimately this research aims to bridge the gap by conducting a comparative study of kernel fat and the impact of geographical diversity on its chemical composition.

Materials and Methods

Sample collection

Mature fruits of sample *M. indica* were collected from different parts of Nepal (Bardiya, Doti, Tanahun, Kathmandu and Dhangadhi) to encapsulate a diverse range of Nepal's ecological zones. These locations represent different altitudes and climatic conditions from Tarai plain to the mid hills. Collected samples were identified by the botanist at the Central Department of Botany, Kirtipur, Kathmandu, Nepal. The collected samples were first washed, chopped, de-pulped, and again the kernel was chopped into fine pieces, sun-dried, and crushed into fine powder. The obtained kernel powder was kept in air-tight container for further use.

Extraction of oil from kernel powder

In the thimble of the Soxhlet apparatus, 60 gm of coarse powder was suspended, and about 300 mL of n-hexane was poured into the round-bottom flask. The Soxhlet apparatus was set up, heated at 70°C, and allowed to stay for 8 hours under continuous extraction. At the end of the extraction, the resulting mixture containing the oil was distilled off to recover the solvent from the oil. The solvent was evaporated using a rotary vacuum evaporator and then dried in an oven at 45°C for 2 hours. The extracted fat was weighed and stored at 20°C until further analysis. The total yield obtained is expressed in percentage by using the following equation to determine the total fat yield of 100 gm of kernel powder on a dry weight basis and was expressed as a percentage:

$$Total\ fat\ yield = \frac{mass\ of\ extracted\ kernel\ fat}{Mass\ of\ mango\ kernel\ powder} \times 100$$

GC-MS analysis of mango kernel butter

The Gas Chromatography-Mass Spectrometer (GC-MS) analysis of mango kernel butter was performed at the Department of Food Technology and Quality Control, Nepal through GC-MS-QP 2010. The column used was RTX5MS and Helium as the carrier gas. The injection temperature was 220°C. The column oven temperature was maintained as follows: hold 80°C for 2 min, followed by an increase to 200°C for 3 min, and then up to 280°C for 4 min and the column head pressure was 67.7 kPa. The sample diluted with spectroscopic grade DCM (Dichloromethane) in a ratio of 1:10 was injected into the GC inlet maintaining a constant flow rate of 1.03 mL min^{-1} and purge flow of 2 mL min^{-1} in split mode using the split ratio of 15.0. The total flow was 18.5 mL min^{-1}. The Mass Spectrometry was performed at the scan speed of 1000 from 3 min up to 30.95 min. Identification of compounds was based on the retention indices determined by comparison of the mass spectral fragmentation patterns with those reported in the NIST 05 library.

Results and Discussion

Properties and percentage yield of sample

Butter was extracted from mango kernel powder collected from five different regions of Nepal using Soxhlet extraction. Among five different sample highest yield was obtained from IM2 (Doti) and lowest yield was obtained from IM4 (Tanahau). This may be due to the difference in moisture content and seed maturity among collected sample. The average percentage yield of the butter from mango kernel sample was calculated to be 7.022% (Table 1). The extracted mango kernel butter, derived from all places, was found to be highly viscous, solid, and sticky with a yellowish-white color.

Fatty acid composition of mango seed butter from different geographical reasons

The GC-MS analysis of mango seed butter showed the presence of varieties of fatty acids (Figure 1). The butter samples extracted from mango seeds from various regions of Nepal showed distinct fatty acid compositions (Table 2**).** The sample IM1 butter extracted from Bardiya district showed five

Table 1: Percentage yield of mango kernel butter

S.N.	Sample	Places	Product	Appearance	Color	Yield (%)	Average Percentage Yield
1	IM1	Bardiya				6.95	
2	IM2	Doti				7.2	
3	IM3	Kathmandu	Butter	Highly Viscous Solid and Sticky	Yellowish white	7	7.02%
4	IM4	Tanahun				6.88	
5	IM5	Dhangadhi				6.98	

Table 2: Fatty acid profile in mango seed butter of different varieties

Compounds (%)	Sample IM1	Sample IM2	Sample IM3	Sample IM4	Sample IM5
Octadecanoic acid	21.97	25.70	12.35	7.76	21.49
l-(+)-Ascorbic acid-2,6-dihexadecanoate	15.60	8.23	-	12.39	8.56
cis-9-Hexadecenal	-	-	52.71	43.68	50.50
2-Isoamyl-6-methyl pyrazine	5.73	2.10	11.48	-	5.13
Oxalic acid, 2-methyl phenyl octadecyl ester	-	5.99	-	-	9.67
6-Octadecenoic acid, (Z)-	47.42	-	-	-	-
Z,Z-4,6-Nonadecadien-1-ol acetate	9.27	-	-	-	-
Z-9-Pentadecenol	-	57.98	-	-	-
n-Hexadecanoic acid	-	-	7.82	-	-
1-Cyclopenta-2,4-dienylundec-10-en-1-one	-	-	15.64	-	-
Octadecanoic acid-2,3-dihydroxypropyl ester	-	-	-	14.91	-
2-Methyl-Z,Z-3,13-octadecadienol	-	-	-	6.07	-
9-Octadecenoic acid (Z)-methyl ester	-	-	-	-	4.65

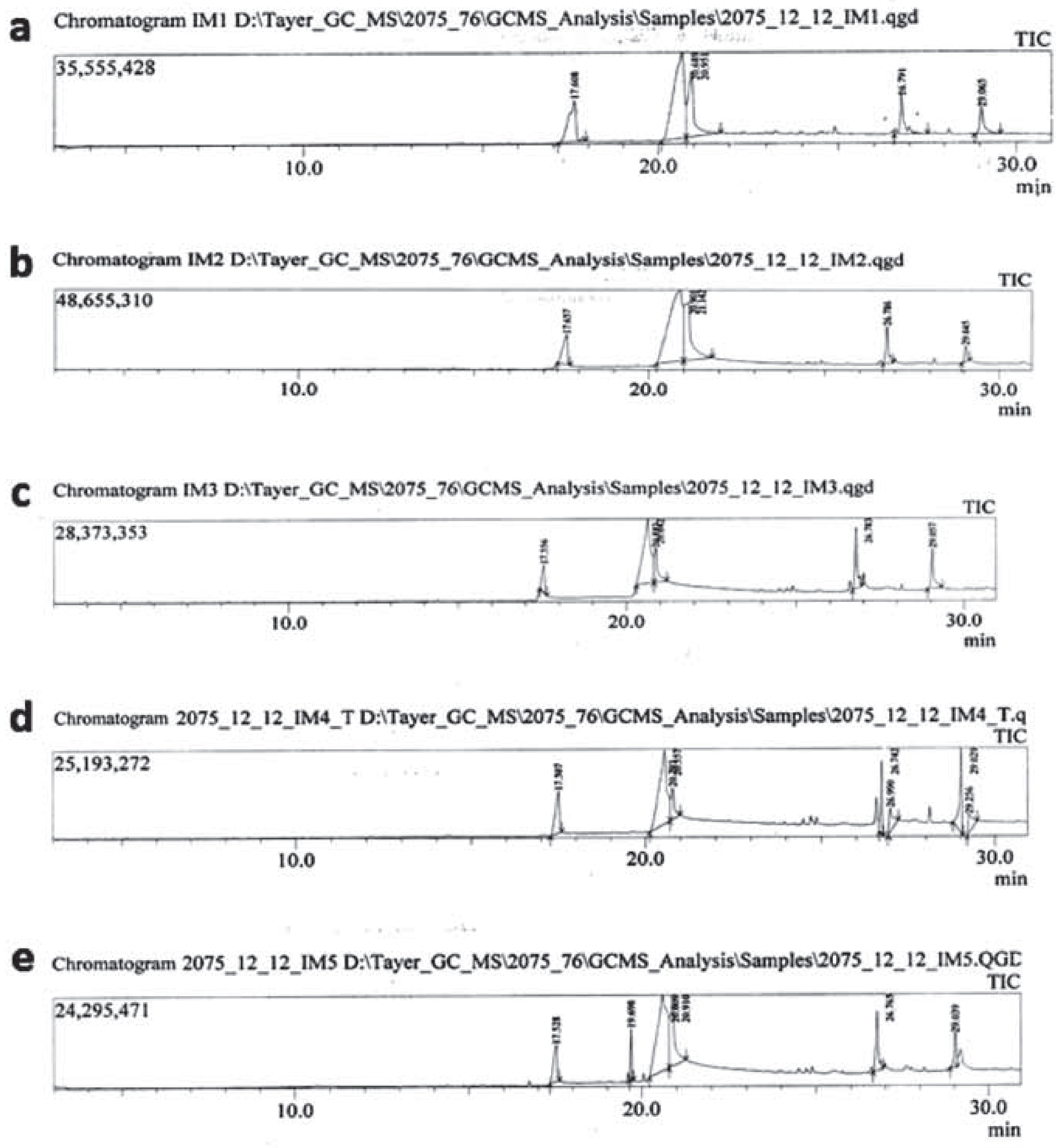

Figure 1: GC-MS chromatogram spectra of mango seed butter samples **(a)** IM1, **(b)** IM2, **(c)** IM3, **(d)** IM4, **(e)** IM5

different compounds with two major fatty acids, 6-octadecenoic acid (47.42%) and octadecanoic acid (21.97%), also known as stearic acid. The sample IM2 butter extracted from Doti showed 5 different compounds with octadecanoic acid i.e. stearic acid (25.70%) as predominant fatty acids. The sample IM3 from Kathmandu showed five different compounds with major fatty acids: n-hexadecanoic acid i.e. palmitic acid (7.82%) and octadecanoic acid i.e. stearic acid (12.35%).

Five different compounds with major fatty acids were octadecanoic acid i.e. stearic acid (25.70%). The sample IM4 from the upper region of Tanahun district in Nepal showed six different compounds with octadecanoic acid (7.76%) as a major fatty acid. The sample IM5 extracted from Dhangadi of Kailali district showed six different compounds with major fatty acids octadecanoic acid i.e. stearic acid (21.49 %) and oleic acid methyl ester i.e. 9- octadecenoic acid methyl ester (4.65%). All the varieties of the mango seed obtained from the different parts of Nepal showed octadecenoic acid but in different ratios. The constituents obtained were similar to those of the other research paper (Muchiri et al., 2012). The variation in the constituents is due to the cultivar, extraction technique, planting area, geographical, altitude, and the many different factors that elevated the constituents in the mango seed (Maldonado-Celis et al., 2019). The primary fatty acids found in mango kernel butter include stearic acid, palmitic acid, and 6-octadecanoic acid, while cocoa butter primarily contains stearic acid, oleic acid, palmitic acid, and linoleic acid (Apgar et al., 1987; Naik & Kumar, 2014). The fatty acid profile of mango kernel butter closely resembles that of commercial cocoa butter. Given the high demand and limited availability of cocoa butter, which contributes to its elevated price, mango kernel butter presents a promising, cost-effective alternative. It offers a high-quality option for replacing cocoa butter in various applications.

Conclusion

Mango (*Mangifera indica* L.) kernels sourced from various districts of Nepal was assessed for their fatty acid composition through GC-MS analysis. The results identified stearic acid, palmitic acid, and octadecanoic acid as the major fatty acid compounds in almost all sample. The difference in the percentage composition may be due to geographical variation and other factors like light, temperature, altitude, tree age, and other factors. Since cocoa butter and mango kernels have similar chemical constituent, mango kernel butter can be a suitable substitute in various application without compromising the desired characteristics of the end product. These findings suggest that mango kernel butter could be a viable and cost-effective alternative to the high-demand and expensive cocoa butter. However, further study must be conducted to know which variety of mango can be best alternative in terms of cocoa butter production, and how geographical condition affects the composition of the butter.

Author Contributions

I Bhatta conceptualized investigations, methodology, and data analysis and prepared the original draft. P Shrestha, P Acharya, S Pantha & R Devkota were involved in preparing the manuscript. A Adhikari helped in language editing.

Acknowledgments

We are thankful to the Department of Food Technology and Quality Control, Nepal for the GC-MS analysis.

Reference

Abdalla, A. E. M., Darwish, S. M., Ayad, E. H. E., & El-Hamahmy, R. M. (2007). Egyptian mango by-product 1. compositional quality of mango seed kernel. *Food Chemistry*, *103*(4), 1134-1140. https://doi.org/10.1016/j.foodchem.2006.10.017

Akhter, S., McDonald, M. A., & Marriott, R. (2016). *Mangifera sylvatica* (wild mango): A new cocoa butter alternative. *Scientific Reports*, *6*, 1-9. https://doi.org/10.1038/srep32050

Anwar, F., Hussain, A. I., Iqbal, S., & Bhanger, M. I. (2007). Enhancement of the oxidative stability of some vegetable oils by blending with *Moringa oleifera* oil. *Food Chemistry*, *103*(4), 1181-1191. https://doi.org/10.1016/j.foodchem.2006.10.023.

Apgar, J. L., Shively, C. A., & Tarka, S. M. (1987). Digestibility of cocoa butter and corn oil and their influence on fatty acid distribution in rats. *The Journal of Nutrition*, *117*(4), 660-665. https://doi.org/10.1093/jn/117.4.660.

Chen, I. S., Subramaniam, S., Vahouny, G. V., Cassidy, M. M., Ikeda, I., & Kritchevsky, D. (1989). A comparison of the digestion and absorption of cocoa butter and palm kernel oil and their effects on cholesterol absorption in rats. *The Journal of Nutrition*, *119*(11), 1569-1573. https://doi.org/10.1093/jn/119.11.1569

Darmawan, D., & Mutalib, A. (2024). Evaluation of environmental impact on cocoa production and processing under life cycle assessment method: From beans to liquor. *Environmental Advances*, *15*, 100481. https://doi.org/10.1016/j.envadv.2023.100481

Kittiphoom, S., & Sutasinee, S. (2013). Mango seed kernel oil and its physicochemical properties. *International Food Research Journal*, *20*(3), 1145-1149.

Lipp, M., Simoneau, C., Ulberth, F., Anklam, E., Crews, C., Brereton, P., De Greyt, W., Schwack, W., & Wiedmaier, C. (2001). Composition of genuine cocoa butter and cocoa butter equivalents. *Journal of Food Composition and Analysis*, *14*(4), 399-408. https://doi.org/10.1006/jfca.2000.0984.

Maldonado-Celis, M. E., Yahia, E. M., Bedoya, R., Landázuri, P., Loango, N., Aguillón, J., Restrepo, B., & Guerrero Ospina, J. C. (2019). Chemical composition of mango (*Mangifera indica* L.) fruit: Nutritional and phytochemical compounds. *Frontiers in Plant Science*, *10*, 1073. https://doi.org/10.3389/fpls.2019.01073

Melo, P. E. F., Silva, A. P. M., Marques, F. P., Ribeiro, P. R. V., Souza Filho, M. de sá M., Brito, E. S., Lima, J. R., & Azeredo, H. M. C. (2019). Antioxidant films from mango kernel components. *Food Hydrocolloids*, *95*(May), 487-495. https://doi.org/10.1016/j.foodhyd.2019.04.061.

Muchiri, D. R., Mahungu, S. M., & Gituanja, S. N. (2012). Studies on mango (*Mangifera indica*, L.) kernel fat of some Kenyan varieties in Meru. *Journal of the American Oil Chemists' Society*, *89*(9), 1567-1575. https://doi.org/10.1007/s11746-012-2054-6

Naeem, A., Shabbir, M. A., Khan, M. R., Ahmad, N., & Roberts, T. H. (2019). Mango seed kernel fat as a cocoa butter substitute suitable for the tropics. *Journal of Food Science*, *84*(6), 1315-1321. https://doi.org/10.1111/1750-3841.14614

Naik, B., & Kumar, V. (2014). Cocoa butter and its alternatives: A review. *Journal of Bioresource Engineering and Technology*, *1*, 7-17.

Shekarchizadeh, H., Kadivar, M., Ghaziaskar, H. S., & Rezayat, M. (2009). Optimization of enzymatic synthesis of cocoa butter analog from camel hump fat in supercritical carbon dioxide by response surface method (RSM). *The Journal of Supercritical Fluids*, *49*(2), 209-215. https://doi.org/10.1016/j.supflu.2009.03.005

Solís-Fuentes, J. A., & Durán-de-Bazúa, M. del C. (2020). Mango seed: Mango (*Mangifera indica L.*) seed and its fats. In V. R. Preedy, & R. R. Watson (Eds.), *Nuts and Seeds in Health and Disease Prevention* (2nd ed.) (pp. 79-90). Academic Press. https://doi.org/10.1016/B978-0-12-818553-7.00008-5

Tharanathan, R. N., Yashoda, H. M., & Prabha, T. N. (2006). Mango (*Mangifera indica L.*), "the king of fruits"- An overview. In *Food Reviews International*, *22*, (2). https://doi.org/10.1080/87559120600574493

Wu, S., Tokuda, M., Kashiwagi, A., Henmi, A., Okada, Y., Tachibana, S., & Nomura, M. (2015). Evaluation of the fatty acid composition of the seeds of *Mangifera indica* L. and their application. *Journal of Oleo Science*, *64*(5), 479-484. https://doi.org/10.5650/jos.ess14238

Journal of Plant Resources (2025) **23**(1), 7-15
https://doi.org/10.3126/bdpr.v23i1.83329

In Vitro Antioxidant and Anti-diabetic Activity of Bark, Flower and Leaf Extracts of *Jacaranda mimosifolia* D. Don

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Abstract

Jacaranda mimosifolia D. Don. (Bignoniaceae) is an ornamental tree with opposite bipinnate leaves and tender lavender-blue flowers. Knowledge of the phytochemicals present in any plant is important for understanding its biological significance which may lead to the discovery of new alternative therapeutic drugs. This paper aims to investigate the phytochemical profile, antioxidant potential and *in vitro* antidiabetic activity of the bark, flowers and leaves of *J. mimosifolia,*extracted using the cold extraction method with various solvents, *viz.* hexane, acetone, 100% ethanol and 70% hydroethanol. Preliminary phytochemical test of all extracts was performed to determine their chemical constituents. The total phenolic content (TPC) and total flavonoid content (TFC) were calculated using the Folin-Ciocalteu method and aluminium chloride colorimetric assay, respectively. Similarly, the *in vitro* antioxidant and antidiabetic potentials of all extracts were determined using the diphenylpicrylhydrazyl (DPPH) assay and α-glucosidase inhibition assay, respectively. The qualitative phytochemical test showed the presence of flavonoids, steroids, glycosides, anthocyanins, terpenoids, tannins and carbohydrates (at least in one part of plant) whereas alkaloids and proteins were completely absent in all the plant extracts. In this study, the highest TPC and TFC were shown in acetone bark extract (219.06 ± 19.54 mg GAE g^{-1}) and hexane leaf (449.76 ± 28.89 mg QE g^{-1}) respectively, whereas the highest IC_{50} for antioxidant activity and anti-diabetic activity were found to be in 100% ethanol bark (56.15 μg mL^{-1}) and hexane flower (92.5% enzyme inhibition), respectively. Due to notable phenolic content, remarkable antioxidant and antidiabetic effects, the phytochemicals present in leaf and flower extracts of *J. mimosifolia* could be considered for the discovery of a new drug against oxidative stress-induced diseases and diabetes.

Keywords: Bignoniaceae, Diphenylpicrylhydrazyl, Medicinal plant, α-glucosidase

Introduction

Since ancient times, plants have been used for therapeutic purposes due to the presence of bioactive compounds and antioxidant properties (Velu et al., 2018). According to the World Health Organization (WHO), approximately 75% of the world's population, especially in developing countries depends on phytotherapeutic agents for primary healthcare (Pan et al., 2014). Phytochemicals have been reported to have various health-promoting effects, including the prevention of the onset of chronic diseases such as cancer, atherosclerosis, neurodegeneration, obesity, articular rheumatism, skin aging and diabetes (Bansal et al., 2021). It has also been reported that they are involved in the pathway of molecular signal transduction, inflammation, defense mechanisms; they can reduce oxidative stress by scavenging deleterious free radicals, or decomposing peroxides, and they help in the regulation of carbohydrate metabolism too (Bansal et al., 2021; Mishra & Tripathi, 2015).

The genus "*Jacaranda*" is an ornamental, woody representative of the Bignoniaceae family having 49 species and found mainly in tropical and subtropical geographical areas (Gachet & Schühly, 2009; Zaghloul et al., 2011). *Jacaranda mimosifolia* D.Don, also called blue jacaranda, black poui, fern tree, is native to Brazil, Bolivia, and Argentina (Xie et al., 2021) and is considered as pioneer tree with opposite bipinnate leaves and tender, deciduous foliage having soft, delicate, dense terminal clusters of lavender-blue, lightly fragrant, trumpet-shaped flowers (El-Marasy et al., 2020; Gilman & Watson, 1993; Mostafa et al., 2014). The pharmaceutical interest of *J. mimosifolia* is increasing due to its richness in bioactive metabolites like alkaloids, flavonoids, polyphenols, iridoid glycosides, tannins, saponins, steroids, triterpene, and acteoside (Adelanwa & Habibu, 2015; El-Marasy et al., 2020; Mostafa et al., 2014).

Traditionally the bark of *J. mimosifolia* had been used to treat venereal diseases like syphilis and to purify the blood in Ecuador (Acosta Solís, 1992; Gachet & Schühly, 2009). The extracts were recorded to treat hypertension, ulcers, wounds, diarrhea, dysentery, amoebic infections in Bangladesh, India and Pakistan, as well as skin problems like, fungal infections, boils, eczema, and psoriasis in Africa(Serra et al., 2020; Sidjui et al., 2014). It possesses antiulcer (Shoukry et al., 2023), antileishmanial, antiprotozoal, anti-inflammatory, and anticancer activities (Naz et al., 2020).Various researchers have found its effective antimicrobial activity against *Bacillus cereus*, *Escherichia coli*, *Salmonella typhii* and *Staphylococcus aureus* (Adelanwa & Habibu, 2015; Aguirre-Becerra et al., 2020). The hydromethanolic extract of leaf was also found to have hypothermic and hypotensive properties via a blockage of alpha-adrenergic receptors (Nicasio & Meckes, 2005; Rahmatullah et al., 2010).

Knowledge of the phytochemicals present in any plant is important for understanding its biological significance which may lead to the discovery of new alternative therapeutic drugs. Though some research has been carried out on phytochemical screening but complete research on various solvent extracts of different parts of *J. mimosifolia* has not been carried out yet. This study aimed to conduct qualitative phytochemical screening, as well as quantitative estimation of total phenolic contents (TPC), total flavonoid contents (TFC) and antioxidant activities with radical scavenging effects of various extracts of *J. mimosifolia* bark, leaves and flower. Also, *in vitro* antidiabetic activity of various parts of *J. mimosifolia* was evaluated.

Materials and Methods

Instruments and chemicals

Rota evaporator from Hahnshin Scientific Co. (Korea) and Bio Tek Epoch-2 Microplate Reader from Biotech (USA) were used. Ascorbic acid, α-p-nitrophenyl α-D-glucopyranoside (nPG) and acarbose were obtained from Sigma- Aldrich (China), 1,1-diphenyl-2-picrylhydrazyl (DPPH) from Sigma- Aldrich (Germany), quercetin from Sigma- Aldrich (India) and gallic acid from Loba Chemie Pvt. Ltd (India). All other chemicals used were of analytical grade.

Collection and extraction of plant materials

The bark, flowers and leaves of *J.mimosifolia* were collected from Thapathali, Kathmandu, Nepal (latitude 27°41'15.28224'' N, longitude 85°19'19.1892" E) at an elevation of 1285 meters above sea level. Then the plant was identified and authenticated at National Herbarium and Plant Laboratories (KATH), Godawari, Lalitpur, Nepal and deposited for future reference. All the laboratory work was carried out at Natural Products Research Laboratory (NPRL), Thapathali, Kathmandu, Nepal. The extracts were prepared by cold extraction method as mentioned by Harborne (1998), Seyfe et al. (2017) and Nortjie et al. (2022). The bark, flowers and leaves were cleaned of extraneous materials, shade dried and ground into fine powder using an electric blender separately. A 200 gm portion of powdered material from each plant part was sequentially macerated at room temperature in 1000 mL of 100% ethanol, followed by 70% ethanol, then acetone, and finally hexane, each for 72 hours with intermittent agitation. Then, the solution was filtered through Whatman grade 1 filter paper, and the residues were macerated further two times with the respective solvents. The filtrates, thus obtained, were concentrated separately using a rotary vacuum evaporator under reduced pressure at 40-45°C to obtain the respective extracts, which were then stored in a refrigerator at 4°C until further use.

Qualitative phytochemical screening

The qualitative phytochemical screenings of twelve extracts of *J. mimosifolia* (bark, flower and leaf extracts obtained using four different solvents) were performed to detect the presence of alkaloids, glycosides, flavonoids, tannins, phenols, saponins, carbohydrates and steroids using the standard methods (Harborne, 1998; Mishra & Tripathi, 2015; Shaikh & Patil, 2020).

Determination of total phenolic contents (TPC) and total flavonoid contents (TFC)

The TPC was determined using the Folin-Ciocalteu's method, taking gallic acid as standard for the calibration curve as described by Singleton and Rossi (1965) with a slight modifications. Briefly, 20 µL of 1 mg mL^{-1} of different extracts was loaded in triplicate in a 96-well plate. Gallic acid at concentrations of 10, 20, 30, 40, 50, 60, 70 and 80 µg mL^{-1} was also loaded in triplicate and used as standard. Then, 100 µL of 10% Folin-Ciocalteu (FC) reagent was added to each well containing either gallic acid or extract. An initial reading of the absorbance was taken at 765 nm using a microplate reader (Epoch2, BioTek, Instruments, Inc, USA). After initial reading, 80 µL of 7% sodium carbonate was added to each well and incubated in dark for 30 minutes. The final absorbance was also taken at 765 nm using the microplate reader. The TPC was expressed in milligrams per gram (mg g^{-1}) of dry plant extract, in gallic acid equivalent (GAE) (Fombang & Mbofung, 2015; Mujic et al., 2009; Seyfe et al., 2017).

Similarly, the TFC were determined by aluminium chloride colorimetric assay, taking quercetin as standard as described by Fombang and Mbofung (2015) and Nobossé et al. (2018). In brief, 20 µL of different concentrations i.e., 10, 20, 40, 60, 80, 100 µg mL^{-1} of quercetin and 20 µL of 1 mg mL^{-1} plant extracts were loaded separately in triplicate in a 96 well plate. Then, 110 µL of distilled water and 60 µL of ethanol were added to each well. The initial absorbance was taken at wavelength 415 nm in a microplate reader. Then, 10 µL of 5% aluminium chloride and 10 µL of 0.5M potassium acetate was added to each plate, followed by incubation in dark for 45 minutes and the final absorbance was taken at the same wavelength i.e., 415 nm. The TFC of each extract was expressed as milligrams of quercetin equivalent per gram of dry weight (mg QEg^{-1}) of the extract.

Evaluation of antioxidant activity

The anti-oxidant activity was evaluated using the DPPH assay, following the method developed by Brand-Williams et al. (1995) with some modifications. Briefly, 10 µL of sample extracts (1 mg mL^{-1}) and ascorbic acid (Vitamin C) of various concentrations as standard were loaded in triplicates in a 96-well plate. Methanol (90 µL) was added and initial absorbance was taken at 517 nm. Then,150 µL of 0.1 mM DPPH solution was added to each well. After incubation for 30 minutes in the dark, absorbance was taken again at 517 nm. The linear plot of percent inhibition versus concentration was analyzed as equation Y = mX + b where, X is the concentration of the measured substance and Y is the percent inhibition. Meanwhile, the IC50 value was determined as the X value of this equation when Y was equal to 50%.

The percentage inhibition of DPPH radicals by the plant extract was calculated using the following formula (Baba & Malik, 2014; Nobossé et al., 2018):

$$\%\ \text{inhibition} = \frac{A_c - A_s}{A_c} * 100$$

Whereas, A_c= absorbance of blank control,
A_s = absorbance of tested sample extract

In vitro antidiabetic assay

The anti-diabetic activity of *J. mimosifolia* extracts was evaluated by α-glucosidase inhibitory activity using α-p-nitrophenyl α-D-glucopyranoside (pNPG) (Sagbo et al., 2018), which is hydrolyzed by α-glucosidase to release p-Nitrophenol (a yellow-colored agent) and acarbose taken as positive standard as mentioned by Bhatia et al. (2019), El Omari et al.(2019) and Marmouzi et al. (2017). For this assay, 20 µL of DMSO as negative standard/

blank, 20 µL of 1 mg mL^{-1} stock solution of plant extract and 20 µL of different concentrations, i.e. 100, 200, 400, 600, 800, 1000 µg mL^{-1} of acarbose were loaded separately in triplicate in a 96-well plate. Then, 20µL of α-glucosidase enzyme (0.5 units mL^{-1}) and 70 µL of phosphate buffer (6.8 pH) were loaded in each plate and left for 15 minutes at 30 °C. The initial absorbance was taken at 405 nm wavelength. Then, 40 µL of pNPG (1.24 mM) was added and reincubated for 30 minutes at 37 °C. The final reading was also taken at 405 nm wavelength after the addition of 50 µL of sodium carbonate (0.2M). The α-glucosidase inhibitory activity was expressed as percentage inhibition.

$$\% \text{ inhibition} = \frac{A_c - A_s}{A_c} * 100$$

Whereas, A_c = absorbance of blank control,
A_s = absorbance of sample extract

Statistical analysis

Data were expressed as mean ± SEM using Microsoft Excel 2021. IBM SPSS (Statistical Package for Social Sciences) version 26 was used for further statistical analyses. One-way ANOVA (Analysis of Variance) test was performed to determine overall mean differences among the various extracts at a 0.05 level of significance (α). Significant ANOVA results were followed by post hoc Dunnett's two-sided test, treating one group as the control and comparing all other extract groups against it.

Results and Discussion

Qualitative phytochemical screening

In this study, flavonoids, steroids, glycosides, anthocyanins, terpenoids, tannins and carbohydrates were detected in the plant extracts (in at least any one part of the plant) whereas alkaloids and proteins were completely absent as shown in Table 1. Naz et al. (2020) also revealed the presence of phenolics, flavonoids, tannins and saponins in all the leaf extract fractions. However, in this study, flavonoids were found in all plant extracts except the acetone and alcohol extracts of bark and the hexane extracts of flowers. Similarly, tannins were found in 70% ethanol extract of leaves and in all flower extracts, except the hexane flower extract. Saponins were detected only in the 70% ethanol extract of flowers and 100% ethanol extract of bark. Previously, methanolic flower extracts of *J. mimosifolia* were found to be rich in phenolics, flavonoids, terpenoids and quinones (Joselin et al., 2013). Additionally, the methanolic leaf extracts have been reported to contain tannins, flavonoids, carbohydrate and to lack triterpenoids and alkaloids (Adelanwa & Habibu, 2015), which is consistent with the findings of our study.

Table 1: Result of phytochemical test of various extracts of *J. mimosifolia*

S.N.	Phytochemicals screening	Methods	70% Hydroethanol			Acetone			Hexane			100% Ethanol		
			Br	Fl	Lf	Br	Fl	Lf	Br	Fl	Lf	Br	Fl	Lf
1	Alkaloids	Hager´s test	-	-	-	-	-	-	-	-	-	-	-	-
		Mayer´s test	-	-	-	-	-	-	-	-	-	-	-	-
2	Flavonoids	NaOH test	++	++	+++	-	+	+	+	-	++	-	+	+
		Shinoda test	++	++	+++	-	+	+	+	-	++	-	+	+
3	Steroids	Salkowski's test	-	-	-	+	+	-	+	+	+++	+	+	-
4	Glycosides	Fehling's test	-	++	-	+	-	-	+	++	+	+	+	-
5	Anthocyanins	Anthocyanin test	++	++	++	++	++	-	-	-	+	+	+	+
6	Terpenoids	Chloroform test	-	-	-	+	+	-	+	+	+	+	+	-
7	Saponin	Froth test	-	+	-	-	-	-	-	-	-	+	-	-
8	Tannins/Phenolics	$FeCl_3$ test	-	++	++	-	++	-	-	-	-	-	+	-
9	Carbohydrates	Molisch test	++	++	++	+	+	+	+	+	+	+	+	+
10	Proteins	Ninhydrin test	-	-	-	-	-	-	-	-	-	-	-	-

Note: + = positive, ++ = mildly positive, +++ = strongly positive, - = negative, Br = bark, Fl = flowers, Lf = leaves

Total phenolic contents (TPC) and total flavonoid contents (TFC)

The quantification of TPC of *J. mimosifolia* revealed that the acetone bark extract had the highest value of TPC (219.06 ± 19.54 mg GAE g^{-1}) followed by 100% ethanol bark (173.32 ± 5.74 mg GAE g^{-1}), while hexane bark extract showed the lowest TPC (19.65 ± 5.4 mg GAE g^{-1}) as shown in Figure 1.The TPC values reported in the present study are higher than the value reported by Aguirre-Becerra et al. (2020) for aqueous flower extract, but lower than the value for methanolic flower extract. In contrast, Rana et al. (2013) found a slightly higher TPC in butanol extracts of leaf and flower than in our study.

Similarly, the quantification of TFC of *J. mimosifolia* showed the absence of flavonoid contents in 70% ethanol extracts of bark and flower, acetone extracts of bark and flower, hexane extract of flower, and 100% ethanol extracts of bark and flower as shown in Figure 1. Flavonoid contents were detected only in the leaf extracts, regardless of whatever solvent was used. Other extracts, except hexane bark extract, lacked flavonoids. TFC was found to be the highest in hexane leaf extract (449.76 ± 28.89 mg QE g^{-1}), while the lowest TFC was observed in 100% ethanol leaf extract (116.42 ± 30.77 mg QE g^{-1}) as shown in Figure 1. Findings are in agreement with TFC value range mentioned by Mostafa et al. (2014). However, Aguirre-Becerra et al. (2020) found higher flavonoid contents even in flower extract while using rutin as standard instead of quercetin.

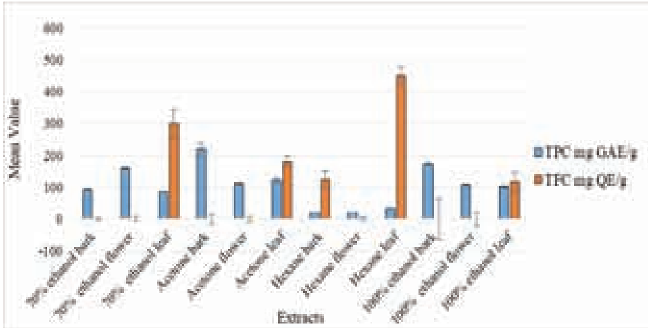

Figure 1: Quantitative value of TPC and TFC of *J. mimosifolia* extracts

Anti-oxidant activity (DPPH Assay)

The DPPH free radical scavenging activity of various extracts of *J. mimosifolia* showed that 100% ethanol bark extract inhibited DPPH free radicals even at low concentration, with an IC_{50} value of 56.15 µg mL^{-1}.This value is slightly lower than that of the ascorbic acid standard (60.26 µg mL^{-1}) as shown in Table 2. Furthermore, the difference between the IC_{50}values of the 100% ethanol bark extract and the positive control (ascorbic acid), was

Table 2: Result of percent inhibition of DPPH free radicals by various extracts of *J. mimosifolia*

Sample	**% of inhibition (Mean±SEM)**			**IC_{50}(µg/mL)**
Concentration (µg/mL)	**25**	**50**	**100**	
Positive control, Ascorbic acid	17.07±0.03	55.02±0.00	73.62±0.01	60.26±0.01
Concentration (µg)	**125**	**250**	**500**	**IC_{50}(µg/mL)**
70% ethanol bark	9.49±0.00	20.21±0.00	41.95±0.00	593.09±0.02
70% ethanol flower	19.56±0.00	43.91±0.00	75.50±0.01	316.83±0.00*
70% ethanol leaf	11.66±0.00	21.15±0.00	42.68±0.00	590.16±0.00*
Acetone bark	26.01±0.00	42.89±0.01	76.08±0.00	304.15±0.00*
Acetone flower	11.88±0.00	25.07±0.00	49.13±0.00	507.12±0.00*
Acetone leaf	20.50±0.00	42.31±0.00	76.30±0.00	316.31±0.00*
Hexane bark	4.61±0.00	6.26±0.00	8.79±0.00	4,245.54±0.00*
Hexane flower	14.23±0.01	15.37±0.01	15.55±0.00	11,493.59±0.01*
Hexane leaf	19.54±0.00	18.65±0.00	20.49±0.00	9,745.34±0.00*
100% ethanol bark	50.85±0.02	68.31±0.01	77.41±0.00	56.15±0.01
100% ethanol flower	33.71±0.00	44.52±0.00	60.08±0.01	348.00±0.00*
100% ethanol leaf	30.92±0.03	44.84±0.03	73.30±0.01	294.44±0.02*

Note: Asterisk (*) indicate statistically significant differences from acarbose at $p < 0.05$ (one-way ANOVA followed by Dunnett's post hoc test)

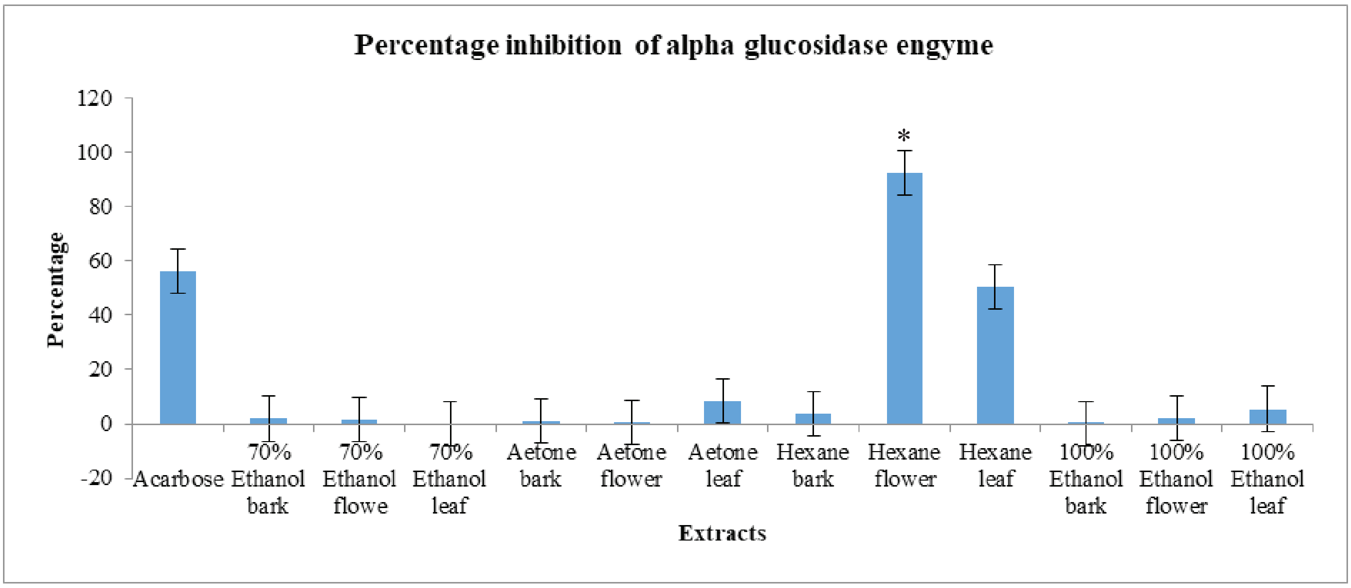

Figure 2: Percentage inhibition of *α*-glucosidase enzyme by *J. mimosifolia* extracts. The values are expressed as mean. Asterisk (*) indicates statistically significant differences from acarbose at $p<0.05$ (one-way ANOVA followed by Dunnett's post hoc test)

found to be insignificant at 5% level of significance, indicating that the antioxidant potential of 100% ethanol bark is comparable to that of ascorbic acid.

In vitro Anti-diabetic assay

The *in-vitro* anti-diabetic assay using the α-glucosidase inhibition test revealed that the enzyme was potently inhibited by hexane flower extract (1 mg mL^{-1}) and hexane leaf extract (1 mg mL^{-1}) of *J. mimosifolia* with inhibition percentage of 92.58 ± 0.19 and 51.47 ± 1.86 respectively (Figure 2). In comparison, the standard used, i.e. acarbose (1 mg mL^{-1}), exhibited an inhibition percentage of 56.21 ± 1.20. Moreover, the hexane flower extract exhibited a statistically significant difference when compared to the positive control, acarbose ($p < 0.05$), indicating a greater α-glucosidase inhibition capacity. In contrast, the hexane leaf extract did not show a statistically significant difference when compared with acarbose ($p = 0.232$), suggesting that its α-glucosidase inhibition capacity is comparable to that of the standard. Our findings are in agreement with those of Serra et.al (2020), who reported that acteoside (verbascoside), isolated from the *J. mimosifolia* leaves, had significant antidiabetic effects in a rat model of type 2 diabetes induced by streptozotocin-nicotinamide (STZ-NA).

Conclusion

Jacaranda mimosifolia showed strong positive reactions for several phytochemical groups like flavonoids, steroids, glycosides, anthocyanin, terpenoids and phenolics, indicating their potential presence in notable amounts. The highest TPC and TFC were shown in acetone bark extract (219.06 ± 19.54 mg GAE g^{-1}) and hexane leaf extract (449.76 ± 28.89 mg QE g^{-1}) respectively whereas the highest DPPH radical scavenging activity and anti-diabetic activity were found to be in 100% ethanol bark extract (56.15 µg mL^{-1}) and hexane flower extract (92.5% inhibition of *α*-glucosidase) respectively. All parts (bark, leaves and flower) of this plant have higher potential for discovery of new drugs. Due to notable phenolic content, remarkable antioxidant and antidiabetic effects, the leaves and flower extracts of *J. mimosifolia* could be considered for further studies on preventing oxidative stress induced diseases and treating diabetes. Therefore, isolation and purification of phytochemicals like jacaranone and acteoside responsible for antioxidant and antidiabetic properties respectively from various parts of *J. mimosifolia* are recommended for discovery of new drug against oxidative stress induced diseases and diabetes.

Author Contributions

All authors have contributed equally to bring the manuscript in this form.

Acknowledgements

We are indebted to National Herbarium and Plant Laboratories (KATH), Lalitpur for taxonomic identification of the herbarium specimens. We extend our deepest gratitude to Mr. Parasmani Yadav from Department of Plant Resources, and Mr. Santosh Basnet, Ms. Monika Bhattarai, Mr. Karma Lal Singh, Mr. Dipak Kumar Shrestha and Mr. Janak Bahadur Lama from NPRL for their kind help and invaluable support during this work.

References

Acosta Solís, M. (1992). *Vademecum de plantas medicinales del Ecuador* [*A handbook of medicinal plants of Equador*]. Fundación Ecuatoriana de Estudios Sociales.

Adelanwa, E. B., & Habibu, I. (2015). Phytochemical screening and antimicrobial activities of the methanolic leaf extract of *Jacaranda mimosifolia* D.Don and *Sansevieria liberica* Thunb. *Journal of Tropical Biosciences, 10,* 1-6.

Aguirre-Becerra, H., Pineda-Nieto, S. A., García-Trejo, J. F., Guevara-González, R. G., Feregrino-Pérez, A. A., Álvarez-Mayorga, B. L., & Pastrana, D. M. R. (2020). Jacaranda flower (*Jacaranda mimosifolia*) as an alternative for antioxidant and antimicrobial use. *Heliyon*, *6*(12), e05802. https://doi.org/10.1016/j.heliyon.2020.e05802

Baba, S. A., & Malik, S. A. (2014). Evaluation of antioxidant and antibacterial activity of methanolic extracts of *Gentiana kurroo* Royle. *Saudi Journal of Biological Sciences*, *21*(5), 493-498. https://doi.org/10.1016/j.sjbs.2014.06.004

Bansal, A., & Priyadarsini, C. (2021). Medicinal properties of phytochemicals and their production. In H. A. El-Shamy (Ed.), *Natural drugs from plants*. IntechOpen. https://doi.org/10.5772/intechopen.98888

Bhatia, A., Singh, B., Arora, R., & Arora, S. (2019). *In vitro* evaluation of the α-glucosidase inhibitory potential of methanolic extracts of traditionally used antidiabetic plants. *BMC Complementary and Alternative Medicine, 19*(1), 74. https://doi.org/10.1186/s12906-019-2482-z

Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. *LWT - Food Science and Technology*, *28*(1), 25-30. https://doi.org/10.1016/S0023-6438(95)80008-5

El Omari, N., Sayah, K., Fettach, S., El Blidi, O., Bouyahya, A., Faouzi, M. E. A., Kamal, R., & Barkiyou, M. (2019). Evaluation of *in vitro* antioxidant and antidiabetic activities of *Aristolochia longa* extracts. *Evidence-Based Complementary and Alternative Medicine, 2019*, 7384735. https://doi.org/10.1155/2019/7384735

El-Marasy, S. A., El-Shenawy, S. M., Moharram, F. A., & El-Sherbeeny, N. A. (2020). Antidiabetic and antioxidant effects of acteoside from *Jacaranda mimosifolia* family biognoniaceae in streptozotocin–nicotinamide induced diabetes in rats. *Open Access Macedonian Journal of Medical Sciences*, *8*(A), 125-133. https://doi.org/10.3889/oamjms.2020.3325

Fombang, E. N., & Mbofung, C. M. F. (2015). Particular benefits can be attributed to *Moringa oleifera* Lam leaves based on origin and stage of maturity. *Journal of Experimental Biology and Agricultural Sciences*, *3*(6), 541-555. https://doi.org/10.18006/2015.3(6).541.555

Gachet, M. S., & Schühly, W. (2009). *Jacaranda*-An ethnopharmacological and phytochemical review. *Journal of Ethnopharmacology*, *121*(1), 14-27. https://doi.org/10.1016/j.jep.2008.10.015

Gilman, E. F., & Watson, D. G. (1993). *Jacaranda mimosifolia*. Fact Sheet ST-317. Environmental Horticulture Department, University of Florida. https://hort.ifas.ufl.edu/database/documents/pdf/tree_fact_sheets/jacmima.pdf

Harborne, A. J. (1998). *Phytochemical methods: A* *guide to modern techniques of plant analysis*. Chapman & Hall Publications.

Joselin, J., Brintha, T. S. S., Florence, A. R., & Jeeva, S. (2013). Phytochemical evaluation of Bignoniaceae flowers. *Journal of Chemical and Pharmaceutical Research*, *5*, 106-111.

Marmouzi, I., Karym, E. M., Saidi, N., Meddah, B., Kharbach, M., Masrar, A., Bouabdellah, M., Chabraoui, L., El Allali, K., Cherrah, Y., & Faouzi, M. E. A. (2017). *In vitro* and *in vivo* antioxidant and anti-hyperglycemic activities of Moroccan oat cultivars. *Antioxidants*, *6*(4), 102. https://doi.org/10.3390/antiox6040102

Mishra, C., & Tripathi, I. (2015). Phytochemical screening of some medicinal plants of Chitrakoot region. *Indian Journal of Applied Research*, *5*(12), 56-60.

Mostafa, N. M., Eldahshan, O. A., & Singab, A. N. (2014). The genus *Jacaranda* (Bignoniaceae): An updated review. *Pharmacognosy Communications*, *4*(3), 1-9.

Mujic, I., Duda , S., Zivkovic, J., Tomin, J., Zekovic, Z., Alibabic, V., Jukiζ, H., & ahinovic, R. (2009). Determination of total phenolic compounds in cultivars of *Castanea Sativa* Mill. *Acta Horticulturae*, *815*, 63-68. https://doi.org/10.17660/ActaHortic.2009.815.8

Naz, R., Roberts, T. H., Bano, A., Nosheen, A., Yasmin, H., Hassan, M. N., Keyani, R., Ullah, S., Khan, W., & Anwar, Z. (2020). GC-MS analysis, antimicrobial, antioxidant, antilipoxygenase and cytotoxic activities of *Jacaranda mimosifolia* methanol leaf extracts and fractions. *PloS One*, *15*(7), e0236319. https://doi.org/10.1371/journal.pone.0236319

Nicasio, P., & Meckes, M. (2005). Hypotensive effect of the hydroalcoholic extract from *Jacaranda mimosaefolia* leaves in rats. *Journal of Ethnopharmacology*, *97*(2), 301-304. https://doi.org/10.1016/j.jep.2004.11.017

Nobossé, P., Fombang, E. N., & Mbofung, C. M. F. (2018). Effects of age and extraction solvent on phytochemical content and antioxidant activity of fresh *Moringa oleifera* L. leaves. *Food Science & Nutrition*, *6*(8), 2188-2198. https://doi.org/10.1002/fsn3.783

Nortjie, E., Basitere, M., Moyo, D., & Nyamukamba, P. (2022). Extraction methods, quantitative and qualitative phytochemical screening of medicinal plants for antimicrobial textiles: A review. *Plants*, *11*(15), 2011. https://doi.org/10.3390/plants11152011

Pan, S.-Y., Litscher, G., Gao, S.-H., Zhou, S.-F., Yu, Z.-L., Chen, H.-Q., Zhang, S.-F., Tang, M.-K., Sun, J.-N., & Ko, K.-M. (2014). Historical perspective of traditional indigenous medical practices: The current renaissance and conservation of herbal resources. *Evidence-Based Complementary and Alternative Medicine*, *2014*(1), 1-20. https://doi.org/10.1155/2014/525340

Rahmatullah, M., Samarrai, W., Jahan, R., Rahman, S., Sharmin, N., Miajee, Z., Chowdhury, M., Bari, S., Jamal, F., Bashar, A., Azad, A., & Ahsan, S. (2010). An ethnomedicinal, pharmacological and phytochemical review of some Bignoniaceae family plants and a description of Bignoniaceae plants in folk medicinal uses in Bangladesh. *Advances in Natural and Applied Sciences*, *4*(3), 236-256.

Rana, A., Bhangalia, S., & Singh, H. P. (2013). A new phenylethanoid glucoside from *Jacaranda mimosifolia*. *Natural Product Research*, *27*(13), 1167-1173. https://doi.org/10.1080/14786419.2012.717290

Sagbo, I. J., van de Venter, M., Koekemoer, T., & Bradley, G. (2018). *In vitro* antidiabetic activity and mechanism of action of *Brachylaena elliptica* (Thunb.) DC. *Evidence-Based Complementary and Alternative Medicine*, *2018*(1), 1-13 https://doi.org/10.1155/2018/4170372

Serra, M. B., Barroso, W. A., Filho, F. F. S., Silva, S. N., Borges, A. C. R., Campos, M. B., Gondim, R. S. D., Souza, R. G., Alves, A. C., Abreu, I. C., & Borges, M. O. R. (2020). Pharmacological evidence from plants of genus *Jacaranda*.

Biomedical Journal of Scientific & Technical Research, *28*(4), 21730-21734. https://doi.org/10.26717/BJSTR.2020.28.004671

Seyfe, S., Toma, A., Esaiyas, A., Debela, E., Fikru, A., & Eyado, A. (2017). Phytochemical screening and *in vivo* antimalarial activities of crude extracts of *Lantana trifolia* root and *Premna oligotricha* leaves in *Plasmodium berghei* infected mice. *Journal of Medicinal Plants Research*, *11*(47), 763-769. https://doi.org/10.5897/JMPR2017.6519

Shaikh, J. R., & Patil, M. (2020). Qualitative tests for preliminary phytochemical screening: An overview. *International Journal of Chemical Studies*, *8*(2), 603-608. https://doi.org/10.22271/chemi.2020.v8.i2i.8834

Shoukry, S. M., El-Hawiet, A., El-Mezayen, N. S., Ghazy, N. M., & Ibrahim, R. S. (2023). Unraveling putative antiulcer phytoconstituents against *Helicobacter pylori* urease and human H+/K+-ATPase from *Jacaranda mimosifolia* using UPLC-MS/MS coupled to chemometrics and molecular docking. *Microchemical Journal*, *189*, 108550. https://doi.org/10.1016/j.microc.2023.108550

Sidjui, L., Menkem, E., Toghueo, R., Noté, O., Mahiou-Leddet, V., Herbette, G., Boyom, F., Ollivier, E., & Folefoc, G. (2014). Secondary metabolites from *Jacaranda mimosifolia* and *Kigelia africana* (Bignoniaceae) and their anticandidal activity. *Records of Natural Products*, *8*(3), 307-311.

Singleton, V. L., & Rossi, J. A. (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American Journal of Enology and Viticulture*, *16*(3), 144-158. https://doi.org/10.5344/ajev.1965.16.3.144

Velu, G., Palanichamy, V., & Rajan, A. P. (2018). Phytochemical and pharmacological importance of plant secondary metabolites in modern medicine. In S. M. Roopan & G. Madhumitha (Eds.), *Bioorganic phase in natural food: An overview* (pp. 135-156). Springer International Publishing. https://doi.org/10.1007/978-3-319-74210-6_8

Xie, C., Zhang, G., Jim, C., Liu, X., Zhang, P., Qiu, J., & Liu, D. (2021). Bioclimatic suitability of actual and potential cultivation areas for *Jacaranda mimosifolia* in Chinese cities. *Forests*, *12*, 951. https://doi.org/10.3390/f12070951

Zaghloul, A. M., Gohar, A. A., Ahmad, M. M., Baraka, H. N., & El-Bassuony, A. A. (2011). Phenylpropanoids from the stem bark of *Jacaranda mimosaefolia*.*Natural Product Research*, *25*(1), 68-76. https://doi.org/10.1080/14786419.2010.511217

Journal of Plant Resources (2025) **23**(1), 16-30
https://doi.org/10.3126/bdpr.v23i1.83336

Phytotoxicity of *Parthenium hysterophorus* L. and *Xanthium strumarium* L. Extracts on Wheat Seed Germination and Cytotoxicity on Brine Shrimp Nauplii

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Abstract

Parthenium hysterophorus L. and *Xanthium strumarium* L. are invasive alien species found around the world. The purpose of this study is to determine the phytotoxicity of *P. hysterophorus* and *X. stumarium* leaf and root extracts on wheat seedlings and their cytotoxicity on brine shrimp [*Artemia salina* (Linnaeus, 1758)] nauplii. The phytotoxicity of various extracts to seedlings was evaluated using petri dishes lined with moistened filter paper, while the cytotoxicity was determined using the brine shrimp lethality assay. It was found that 85-90% of wheat seeds germinated in dichloromethane (DCM) and methanol extracts of leaf and root of *P. hysterophorus and X. strumarium* at lower concentrations (0.15 to 0.62 mg/mL), 50-60% at medium concentration (1.25 and 2.5 mg/mL), and 10-20% at higher concentration (5 mg/mL) after three days of incubation. Root methanol extracts were more phytotoxic to root and shoot growth in seedlings (at 0.62-5.0 mg/mL) than other *P. hysterophorus* root and leaf extracts. Similarly, root DCM extracts were more phytotoxic to root and shoot growth in seedlings (at 0.31-5.0 mg/mL) than other *X. strumarium* root and leaf extracts. Moreover, the root DCM extract was more cytotoxic (LC_{50}: 302.93 μg/mL) to brine shrimp nauplii than the other extracts of root and leaf of *P. hysterophorus.* Similarly, root DCM extract was more cytotoxic (LC_{50}: 129.16 μg/mL) to brine shrimp nauplii than the other extracts of root and leaf of *X. strumarium.* This study revealed that the allelochemicals/phytotoxins present in the methanol and DCM extracts of the roots and leaves of *P. hysterophorus* and *X. strumarium* were phytotoxic to wheat seeds and seedlings, as well as cytotoxic to brine shrimp nauplii. This work will help future research into the effects of allelochemicals on natural vegetation and other crop plants, as well as the carcinogenic activity of allelochemicals identified in *P. hysterophorus* and *X. strumarium* on different human cell lines.

Keywords: Allelochemicals, Allelopathy, DCM, Extract, Seedling

Introduction

Parthenium hysterophorus L. (Asteraceae; wild carrot weed) and *Xanthium strumarium* L. (Asteraceae; rough cocklebur) are invasive alien species (Invasive Species Specialist Group [ISSG], n.d.). *P. hysterophorus* is distributed in many tropical and subtropical regions of the world, including Asia, Africa, Australia, and the Pacific region (Adkin & Shabbir, 2014), causing threats to natural environments, the agricultural sector, and conservation activities. *P. hysteroporus* contains certain allelochemicals, such as parthenin, hysterin, hymenin, and ambrosin that exert significant allelopathic impacts on different crop plants (Dukpa et al., 2020). In many dicotyledons and monocotyledon plants, the parthenin suppresses seed germination and radicle growth. Similarly, *X. strumarium* is originally native to North and South America; however, it has since become extensively naturalized and may be found primarily in temperate, subtropical, and Mediterranean regions (Waheed et al., 2024). *X. stramurium* contains allelochemicals like 1, 5-Dimethyltetralin, Eudesmol, l-Borneol, Ledene alcohol, -(-) Caryophyllene oxide, Isolongifolene, 7,8-dehydro-8a-hydroxy, L-Bornyl acetate, and Aristolene epoxide. According to Duke and Lydon (1987) *X. strumarium* consists of a variety of secondary metabolites and allelochemicals, such as sesquiterpene lactones (xanthatin and xanthinosin), tannins, flavonoids, and alkaloids. These chemicals, especially the sesquiterpenoids, and monoterpenes, were demonstrated to hinder the growth and germination of other types of plants, including

Bidens pilosa L., in a concentration-dependent way (El-Gawad et al., 2019). Both xanthatin and xanthinosin have potent phytotoxic effects by interfering with target plant cellular functions such as mitochondrial function and membrane stability.

Phytotoxicity refers to the detrimental effects of particular compounds (phytotoxins) or growing circumstances on plants, such as delayed seed germination or inhibited growth, root development, and overall health of plants (REAL CCS, 2014). Phytotoxinsare the toxic substances produced by plants, such as juglone (produced by *Juglans regia L.*), cyanogenic glycosides (produced by *Manihot esculenta* Crantz, *Sorghum bicolor* (L.) Moench, *Prunus avium* (L.) L.), coniine (produced by *Conium maculatum* L.), etc. that causes phytotoxicity. Allelopathy is a biological phenomenon in which organisms, particularly plants, emit biochemicals (allelochemicals) into the surroundings that affect the growth, existence, and reproduction of other living things (Rice, 1984). These compounds can have either inhibitory or stimulatory impacts on plant competitiveness, soil microbial populations and ecosystem dynamics (Cheng & Cheng, 2015). However, allelopathy may be used as natural herbicides for weed management (Weston & Duke, 2003), sustainable agriculture to reduce chemical pesticide dependency (Cheng & Cheng, 2015), and ecosystem dynamics to influence plant community structure and invasive species competition (Inderjit & Duke, 2003). Allelopathic interactions take place through a variety of mechanisms, including root exudation (Weston & Duke, 2003), leaf leachates (Cheng & Cheng, 2015), gas volatilization (He et al., 2012), and plant residue decomposition (Inderjit & Duke, 2003) in nature. Allelopathic compounds frequently impede root and shoot growth in targeted plants by interfering with cell division, enzyme activity, and hormone control (Rice, 1984).

Cytotoxicity is the capacity of a substance to harm or kill live cells. It is an important measure for determining the hazardous potential of natural or synthesized substances, particularly in drug development, environmental toxicology, and research on agriculture (Kroemer et al., 2009). Although cytotoxic compounds are also capable of killing normal human cells, their primary function is to selectively destroy rapidly dividing cells, including cancer cells. Therefore, cytotoxic compounds can be used to make drugs by making them specifically designed to target tumor cells and not normal cells. However, it may not function precisely, and that's why people experience side effects of drugs. Cytotoxicity studies investigate cell viability, integrity of the membrane, metabolism, and induction of apoptosis (Wang et al., 2012). The brine shrimp lethality assay (BSLA) is a popular cytotoxicity testing bioassay that utilizes *Artemia salina* nauplii (larvae) as an experimental organism. This assay is inexpensive, straightforward, and offers preliminary information about the hazardous or bioactive qualities of plant extracts (Meyer et al., 1982).

The allelopathic effect of invasive alien species on natural vegetation has been the subject of several studies; however, the allelovpathic effect of these invasive species on crop yield has received less attention. According to Naderi et al. (2024), *P. hysterophorus* decreased maize yields by as much as 46% at a density of 16 plants/m² according to a two year field research conducted in Pakistan. Even at low densities, losses in yield showed a nonlinear pattern, with notable decreases (e.g., 14% loss at 1 plant/m²). Hussain et al. (2014) studied the effect of *X. strumarium* on maize production. A few studies have been conducted on the effect of growth and productivity of wheat (*Triticum aestivum* L.) caused by *X strumarium* and *P hysterophorus* because of their potential phytotoxic compounds, competition for nutrients, light, and space, and allelopathic effects. Similarly, limited research has been conducted to study the effect of dichloromethane (DCM) and methanol extracts of leaf and root extracts of *P. hysterophorus* and *X. stramurium* on wheat seed germination and cytotoxicity on brine shrimp nauplii in lab condition. In a petri dish experiment, Shakya et al. (2021) studied the effect of *P. hysterophorus* leachates on wheat seed germination and growth metrics. The effect of *P. hysterophorus* residues on specific soil characteristics and the growth of *Cicer arietinum* and *Raphanus sativus* in soil and a lab setting were investigated by Batish et al. (2002).

Zahid et al. (2014) studied the effect of *Xanthium strumarium* on maize yield and yield components under natural soil conditions and found a reduction in the percentage yield of grain.

The majority of the research focused on the influence of methanol and aqueous extracts of leaves of *P. hysterophorus* (Amare, 2018; Bashar et al., 2021; Bashar et al., 2023) and leaves and fruits of *X. stramurium* (Benyas et al., 2010; Jalali et al., 2013; Mirzaee & Saeedipour, 2021; Seifu et al., 2024) on seed germination. A comparison of the allelopathic effect of leaf and root extracts on plants (seed germination) and their cytotoxic effect on animals (brine shrimp nauplii) could bring insight into the physiological link between plants and animals. Therefore, this research aims to assess the effect of allelochemical present in dichloromethane and methanol extracts of *P. hysterophorus* and *X. strumarium* leaf and roots on wheat seed germination, radicle and plumule growth in seedlings and cytotoxic potential in brine shrimp nauplii.

Materials and Methods

Plant collection and identification

P. hysterophorus and X. strumarium were collected in December 2023 at a height of 150 meters above sea level from Butwal, Western Nepal (Figure 1). The collected plants (Voucher no. PH05, XS07) were identified by using standard literature (Banik & Yomso, 2021; Chinnuswamy et al., 2018; Rajbhandari et al., 2016) and deposited in the Department of Botany, Butwal Multiple Campus, Tribhuvan University, Butwal, Nepal.

Shade drying and pulverization

The leaves and roots were cleaned under running tap water, dried in the shade, and then laid on newspaper at room temperature until completely dry. An electric grinder was used to grind the fully dried leaves and roots into a fine powder.

Extract preparation

Leaf and root powders (50 gm each) were macerated in 100 mL methanol and dichloromethane (DCM) separately at room temperature for 48 hours. They were filtered with the use of Whatman No. 1 filter paper. The residue was again macerated with 100 mL methanol and DCM for 24 hours twice more to complete the extraction, and the solution mixture was filtered using filter paper. The filtrate was evaporated and concentrated using a rotary evaporator at 37^{o}C. The samples were then kept at 4˚C in refrigerator for further testing.

Figure 1: Collected plants, **(A)** *Parthenium hysterophorus,* **(B)** *Xanthium strumarium*

Evaluation of wheat seed germination and growth of seedlings using various extracts of P. hysterophorus and X. strumarium

Wheat seed collection and sterilization: Wheat (*Triticum aestivum*) seeds were obtained from the National Wheat Research Program, Bhairahawa, Rupandehi. The seeds were thoroughly rinsed with tap water and soaked in water for 24 hours. The seeds were then sterilized for 10 minutes with 1% sodium hypochlorite before being washed three times with sterilized distilled water. Filter papers, petri dishes, forceps, needles, and distilled water were sterilized in an autoclave at 121^{o}C at 15 lb pressure for 15 min.

Seed germination on petri dishes and seedling growth: Petri dishes were lined with blotting papers at the base and moisturized with sterile distilled water. Sterilized 10 uniform seeds were placed in each petri dish using forceps. Seeds were treated with 5.0, 2.5, 1.25, 0.62, 0.31, and 0.15 mg/mL concentration of extracts by the serial dilution (using sterile distilled water) of 10 mg/mL stock solution of DCM and methanol root and leaf extracts of *P. hysterophorus and X. strumarium*. Seeds treated with sterile distilled water were considered as the control treatment. The entire process was carried out in an air laminar flow cabinet. Now, the petri dishes with seeds were closed with a lid, labeled, and kept inside the incubator at 26±2°C for seed germination. The germination of seeds under different treatments was observed after every three days. The number of germinated seeds was recorded, and the radicle (root) and plumule (shoot) lengths in seedlings treated with concentration of extracts were measured with the help of a scale for 12 days.

Assessment of brine shrimp lethality assay using various extracts of P. hysterophorus and X. strumarium

The cytotoxicity of methanol and DCM crude extracts was assessed using a brine shrimp lethality assay (Fatope et al., 1993; Meyer et al., 1982) with a slightly modified protocol. First, 3.5 gm of NaCl was added and dissolved in 100 mL of distilled water to prepare an artificial seawater solution. Eggs weighing approximately 10 mg of brine shrimp (*Artemia salina*) were incubated in seawater for 48 hours at 23°C (using an 80-watt light bulb) to hatch the eggs into larvae or nauplii. Stock solution (10 mg/mL) of each plant extract (*P. hysterophorus* and *X. strumarium*) was prepared in a 2 mL Eppendorf tube separately. Then, various concentrations (5, 2.5, 1.25, 0.62, 0.31, and 0.15 mg/mL) of each extract were prepared using the serial dilution method with salt water. Potassium dichromate (concentrations of 10, 20, 40, 80, and 160 µg/mL) was employed as the positive control, and seawater (saltwater) as the negative control. In a well plate, a total of 150 µL of different concentrations of extracts and saltwater with 15 brine shrimp nauplii were treated with the help of a micropipette. It was incubated in light for 24 hours, and the number of living nauplii was determined. For every sample, triplicate experiments were conducted. Each plant extract's lethal concentration that causes 50% death (LC_{50}) was calculated using the regression line that was created by graphing the concentration against the percentage of mortality on a probit scale.

$$\% \text{ Mortality} = \frac{\text{No. of dead larvae (Nauplii)}}{\text{Initial No. of live larvae (Nauplii)}} \times 100$$

Data analysis

After three days of culture in petri dishes, the percentage of wheat seed germination with the control and extract treatments was determined using Microsoft Excel 2010.The mean length and standard error of the length of the roots and shoots of wheat seedlings with the control and various extract treatments in seeds were calculated after 3, 6, 9, and 12 days using Microsoft Excel 2010.Similarly, the LC_{50} of brine shrimp nauplii in various extract concentrations was computed using a simple linear regression line, and the percentage mortality of brine shrimp nauplii was determined using Microsoft Excel 2010. The correlation between extracts of *P. hysterophorus* and *X. strumarium* and the growth of wheat seedlings was determined using Principal Component Analysis (PCA) using R-software version 4.3.

Results and Discussion

Evaluation of wheat seed germination using various extracts of P. hysterophorus and X. strumarium

This study showed that wheat seeds were germinated initially into seedlings in various concentrations of leaf and root extracts of *P. hysterophorus and X. strumarium* in petri dish moistened with filter paper; however, the growth and development of radicle (root) and plumule (shoot) in seedlings differed depending on the concentration of extracts and time. Seed germination and growth and development of roots and shoots in wheat seedlings were evaluated for 12 days. It was found that 85-90% of wheat seeds germinated in DCM and methanol extracts of leaf and root of *P. hysterophorus and X. strumarium* at lower concentrations (0.15, 0.31, and 0.62 mg/mL), 50-60% at medium concentration (1.25 and 2.5 mg/mL), and 10-20% at higher concentration (5 mg/mL) after three days of incubation (Tables 1 and 2). This study also showed that DCM and methanol extracts from both plants had more or less similar effects on seed germination after three days, however, compared to *P. hysterophorus*, a somewhat lower percentage of seeds germinated in the DCM and methanol extracts of *X. strumarium* (Tables 1 and 2). Several studies showed that *X. strumarium* contains xanthatin and xanthinin, which prevent various kinds of crops and weeds from germinating and growing, indicating a potential allelopathic function. Shajie and Saffari (2007) found that *X. strumarium* leaf and stem extracts significantly lowered the germination, root, and shoot lengths of all tested crops, including corn, canola, sesame, lentils, and chickpea. Though seeds may not have had permanent physiological alterations within three days due to the application of extracts, the germination effect appeared in seeds. Slow penetration and bioaccumulation of lipophilic extracts (like DCM) and polar metabolites (like methanol-extracted phenolics) may be the cause of the delayed physiological response in seeds. These extracts may also take longer to pass through seed coats or cellular membranes. Plant extracts containing phenolic acids, such as ferulic acid, impair mitochondrial function and membrane integrity; however, because of their slow absorption, their effects require time to become visible (Einhellig, 1994). However, unlike soil, where factors such as microbial interactions, availability of nutrients, and retention of water change, a filter paper setup ensures consistent moisture delivery

Table 1: Percentage of seed germination in DCM and Methanol extracts of Leaf and root of *P. hysterophorus* after the three days

Extract concentration (mg/mL)	Total no. of seeds	Average no. of germinated seeds	Percentage of seed germination
0.15	120	108.00	90.00
0.31	120	106.85	89.04
0.62	120	104.57	87.14
1.25	120	72.00	60.00
2.5	120	66.57	55.47
5.0	120	24.28	20.23

Table 2: Percentage of seed germination in DCM and Methanol extracts of Leaf and root of *X. strumarium* after the three days

Extract concentration (mg/mL)	Total no. of seeds	Average no. of germinated seeds	Percentage of seed germination
0.15	120	105.42	87.85
0.31	120	103.42	86.18
0.62	120	102.28	85.23
1.25	120	68.28	56.90
2.5	120	59.71	49.75
5.0	120	12.00	10.00

and similar exposure to plant extracts, minimizing external variation (Khan et al., 2011). Some allelopathic chemical substances can affect seed coat permeability, reducing water absorption and slowing or entirely suppressing germination (Chou, 1999). Similarly, several allelochemicals found in plant extracts suppress amylase and protease enzymes, depriving the embryo of vital nutrients (Li et al., 2010). This technique enables a direct assessment of germination responses to various extract concentrations, including possible inhibiting or stimulating effects (Bewley et al., 2013).

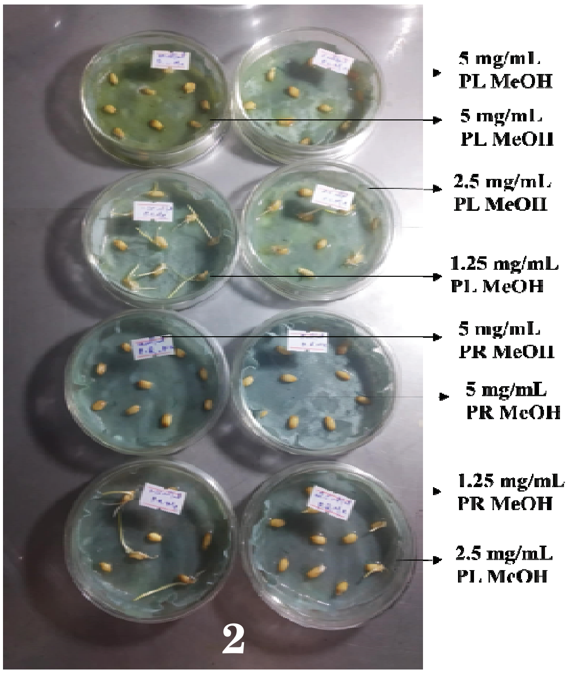
Effect of DCM and methanol extracts of P. hysterophorus on the growth and development of shoots and roots in wheat seedlings

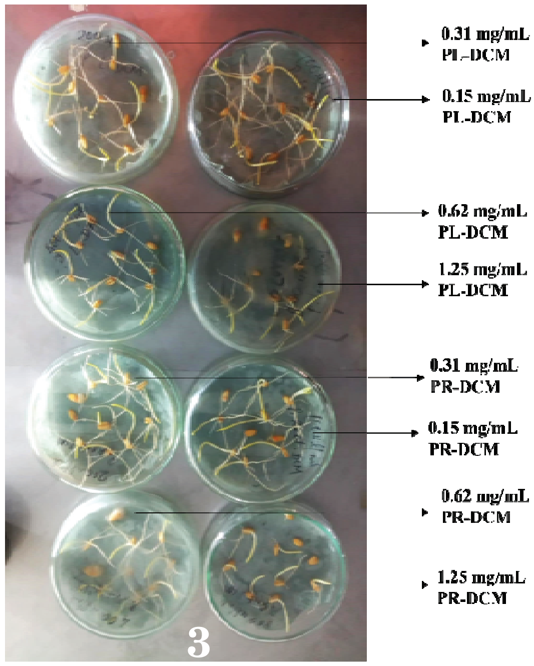
This study showed that methanol extracts of leaf and roots were more effective for the growth inhibition of root and shoots in wheat seedlings than the DCM extracts of leaf and roots of *P. hysterophorus* (Tables 3 and 4; Figures 2 and 3). Methanol is a polar solvent that may extract a wider variety of allelochemicals and phytotoxins, such as phenolics, flavonoids, tannins, alkaloids, phenolic acids (e.g., ferulic acid, p-coumaric acid), and parthenin (Harborne, 1998; Singh et al., 2002). These compounds inhibit cell division, activity of enzymes, and uptake of nutrients, resulting in decreased root and shoot growth (Singh et al., 2003). According to studies, methanol extracts of *P. hysterophorus* have higher amounts of water-soluble allelochemicals that affect the seedling metabolism (Kanchan & Jayachandra, 1980). The growth inhibition of roots and shoots in seedlings was directly proportional to extract concentrations, while the growth and development of roots and shoots in seedlings were found to be directly proportional to time. During the initial growth stage of seedlings for three days, the length of roots and shoots was greater in lower concentrations (from 0.15 to 0.62 mg/L) of DCM and methanol extracts of leaf and root than in the control treatment; however, at higher concentrations, the length of roots and shoots were decreased than that of the control treatment (Tables 3 and 4). It may be due to hormesis, which is an alternating response in which low concentrations of hazardous chemicals stimulate growth while greater quantities inhibit it (Calabrese & Baldwin, 2003). At low concentrations (0.15 - 0.62 mg/L), some phenolic compounds and sesquiterpene lactones of *P. hysterophorus* might act similarly to plant growth regulators.

When compared to the extracts of plant parts, root extracts (both methanol and DCM) of *P. hysterophorus* showed a higher inhibition for the growth and development of roots and shoots in wheat seedlings than the leaf extracts (both methanol and DCM). It could be owing to the larger concentration of allelochemicals in roots than in leaves. In nature, plants discharge allelochemicals into the soil through their roots. This process can happen actively during root growth or passively via diffusion. The discharges may contain phenolics, flavonoids, terpenoids, and organic acids, all of which may interfere with seed germination or plant growth (Inderjit & Duke, 2003).

Table 3: Effect of DCM and methanol leaf extracts of *P. hysterophorus*on wheat seed germination

Extracts	Concentration (mg/mL)	Mean length of roots ± S.E (cm)				Mean length of shoots ± S.E (cm)			
		3 days	6 days	9 days	12 days	3 days	6 days	9 days	12 days
DCM	5.0	0.3±0.51	0.6±0.35	0.8±0.67	0.8±0.56	0.5±0.65	0.8±0.49	1.2±0.56	2.5±0.45
	2.5	0.3±0.67	1.3±0.67	1.5±0.45	2.1±1.12	0.4±0.87	0.9±0.41	2.5±67	3.4±0.76
	1.25	0.4±0.54	2.3±0.98	2.4±0.98	2.4±1.54	0.6±0.45	4.6±1.64	8.9±2.42	9.6±1.54
	0.62	1.6±0.87	3.7±0.54	4.4±0.45	5.5±0.54	0.6±0.31	5.2±2.21	10.6±2.65	11.5±4.21
	0.31	1.6±0.39	3.95±0.71	4.6±1.21	4.6±0.43	0.7±0.12	4.5±3.11	9.8±3.21	12.5±2.54
	0.15	2.5±0.56	4.8±1.1	5.6±0.62	6.1±2.21	1.2±0.54	5.4±0.78	10.7±4.42	12.3±1.87
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54
Methanol	5.0	0.1±0.43	0.1±0.56	0.1±0.78	0.4±0.34	0.4±0.54	0.7±0.54	0.8±0.64	1.2±0.76
	2.5	0.2±0.34	0.6±0.51	0.8±0.34	1±0.76	0.2±0.34	0.9±0.76	2.1±0.34	2.3±0.52
	1.25	0.4±0.87	1.1±0.86	1.3±0.87	1.5±0.34	0.5±0.52	1.4±1.23	1.6±0.44	1.9±0.76
	0.62	1.1±0.21	2.8±0.42	3.8±1.42	4.5±2.21	0.7±0.76	1.8±0.67	4.4±1.54	5.2±1.54
	0.31	1.5±0.67	3.3±0.47	4.2±2.41	5.2±1.65	0.7±0.34	3.1±2.41	7.8±2.54	9.9±3.45
	0.15	2.2±0.51	4.1±1.31	4.7±1.52	6.2±1.65	1.3±0.87	3.9±1.87	8.3±3.31	10.1±0.8
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54





Figures 2 and 3: Effect of methanol and DCM extracts in wheat seedlings at various concentrations (PR MeOH=*Parthenium* root methanol extract, PL MeOH= *Parthenium* leaf methanol extract, PR-DCM= *Parthenium* root DCM extract, and PL-DCM= *Parthenium* leaf DCM extract)

Effect of DCM and methanol extracts of X. strumarium on the growth and development of shoots and roots in wheat seedlings

The growth inhibition, as well as the growth and development of roots and shoots in seedlings, was shown to be directly proportional to extract concentrations and time. This study showed that DCM extracts of leaf and roots were more effective for the growth inhibition of roots and shoots in seedlings than the methanol extracts of leaf and roots of *X. strumarium* (Tables 5 and 6; Figures 4 and 5). DCM is more effective at separating non-polar and semi-polar compounds, such as terpenoids, steroids, and specific flavonoids, that are frequently linked to allelopathic and phytotoxic actions (Gniazdowska & Bogatek, 2005). DCM-extracted non-polar compounds may permeate plant cell membranes more effectively, causing greater toxicity and impairment of cellular functions than polar methanol extracts (Macías et al., 2007). According to Singh et al. (2003) extracts of *X. strumarium* produced using non-polar solvents had more biological activity due

Table 4: Effect of DCM and Methanol root extracts of *P. hysterophorus* on wheat seed germination

Extracts	Concentrations (mg/mL)	Mean length of roots ±S.E (cm)				Mean length of shoots ±S.E (cm)			
		3 days	6 days	9 days	12 days	3 days	6 days	9 days	12 days
DCM	5.0	0.2±0.34	0.7±0.88	0.9±0.34	1.1±0.56	0.2±0.12	1.2±0.67	3.3±0.38	4.4±0.56
	2.5	0.4±0.56	0.8±0.56	1.2±0.78	1.8±0.53	0.3±0.34	1.2±0.51	4.5±0.81	5.1±0.34
	1.25	0.4±0.67	2.1±0.23	2.3±1.57	2.5±86	0.4±0.54	3.5±1.09	8.5±2.23	9.3±2.03
	0.62	1.8±0.83	4.1±1.04	4.4±2/09	4.6±1.85	0.7±0.32	4.8±2.65	10.1±0.4	11.5±0.6
	0.31	1.9±0.56	4.5±2.13	4.8±0.52	5.1±2.54	0.7±0.31	4.4±1.52	8.3±0.75	10.1±0.8
	0.15	1.9±0.76	4.9±0.89	5.1±0.62	5.2±1.37	0.9±0.46	5.5±2.75	10.2±0.8	11.2±0.4
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54
Methanol	5.0	0.0±0.00	0.0±0.00	0.0 ±0.0	0.0±0.0	0.0±0.00	0.0±0.00	0.0±0.0	0.0±0.0
	2.5	0.1±0.44	0.2±0.34	0.2±0.43	0.3±0.34	0.1±0.45	0.4±0.23	1.4±0.21	2.0±0.34
	1.25	0.2±0.33	0.4±0.35	1.7±0.53	2.3±0.76	0.3±0.54	1.6±0.35	6.4±0.53	9.5±0.76
	0.62	0.3±0.56	0.8±0.56	2.3±0.76	3.4±0.65	0.4±0.32	2.1±0.76	6.8±0.61	9.5±1.23
	0.31	0.7±0.62	2.5±0.66	3.9±0.91	4.2±1.05	0.6±0.23	2.8±0.61	7.9±1.74	10.1±0.8
	0.15	0.8±0.23	2.9±1.05	4.1±1.03	4.5±0.54	0.6±0.34	3.3±0.67	8.3±2.34	10.5±0.6
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54

Table 5: Effect of DCM and Methanol leaf extracts of *X. strumarium*on wheat seed germination

Extracts	Concentrations (mg/mL)	Mean length of roots ±S.E (cm)				Mean length of shoots ±S.E (cm)			
		3 days	6 days	9 days	12 days	3 days	6 days	9 days	12 days
DCM	5.0	0.0±0.0	0.1±0.21	0.1±0.31	0.2±0.21	0.0±0.0	0.3±0.23	0.5±0.23	0.8±0.45
	2.5	0.1±0.23	0.1±0.34	0.3±0.21	0.4±0.23	0.1±0.23	0.4±0.34	0.9±0.45	2.3±0.54
	1.25	0.4±0.32	0.9±0.54	1.3±0.54	1.4±0.51	0.1±0.23	1.1±0.56	2.4±0.54	5.7±0.89
	0.62	0.5±0.32	1.0±0.23	1.5±0.76	1.9±0.48	0.2±0.32	1.4±0.54	3.6±0.78	6.3±1.29
	0.31	0.5±0.23	1.5±0.76	2.2±1.09	3.1±1.23	0.2±0.23	1.7±0.76	5.3±1.06	7.1±2.92
	0.15	0.8±0.54	3.1±0.32	3.9±0.89	4.5±2.14	0.3±0.21	2.3±97	8.2±1.38	10.2±0.5
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54
Methanol	5.0	0.1±0.24	0.1±0.23	0.1±0.23	0.1±0.23	0.1±0.32	0.1±0.23	0.7±0.34	1.4±0.43
	2.5	0.2±0.26	0.3±0.32	0.4±0.41	0.5±0.23	0.1±0.23	0.5±0.34	1.9±0.76	3.5±0.49
	1.25	0.6±0.43	1.3±0.45	1.3±0.76	1.5±0.65	0.2±0.34	1.6v0.65	4.5±0.87	5.6±0.76
	0.62	0.8±0/49	1.5±0.57	1.8±0.87	3.5±0.76	0.3±0.24	1.8±0.45	5.1±0.56	6.5±1.04
	0.31	1.1±0.78	3.1±0.78	3.6±0.56	4.8±1.92	0.4±0.31	2.7v0.78	8.3±0.54	10.2±0.6
	0.15	1.3±0.36	3.8±0.87	4.2±0.67	5.3±0.34	0.5±0.42	3.8±0.34	9.7±0.87	12.5±0.8
	Control	0.6±0.87	2.8±0.54	3.6±0.76	4.1±0.94	0.3±0.56	2.6±0.56	7.7±0.78	9.8±0.54

to the presence of lipophilic secondary metabolites. During the initial growth stage of seedlings for three days, the length of roots and shoots was greater in lower concentrations (from 0.15 to 0.31 mg/L) of DCM and methanol extracts of leaf and root than in the control treatment; however, at higher concentrations, the length of roots and shoots were decreased than that of the control treatment as in *P. hysterophorus* (Tables 5 and 6).

When compared to the extracts of plant parts, root extracts (both methanol and DCM) of *X. strumarium* showed a higher inhibition for the growth and development of roots and shoots in wheat seedlings than the leaf extracts (both methanol and DCM). It might be because roots have a higher concentration of allelochemicals than leaves do. However, allelochemicals enter the soil via many mechanisms and affect plant competition by reducing seed germination, root elongation, and shoot elongation in nature. The longevity and impact of these compounds are determined by soil composition, microbial activity, and environmental conditions.

Correlation between extracts of P. hysterophorus and X. strumarium and growth of wheat seedlings using principal component analysis (PCA)

The effect of plant extracts treatment of *Parthenium hysterophorus* and *Xanthium strumarium* on the growth of shoot and roots in wheat seedling was analyzed by PCA (Appendix 1 & 2). The result of the individual showed that the proportion of variance was found in axes PCA1 to PCA%. However, more than 95% of the proportion of variance was explained by PCA1 and PCA2.

The biplot of PCA on DCM and Methanol leaf extract (Figure 6) and root extract (Figure 7) of *P. hysterophorus* showed negative relationships with shoot and root elongation of wheat seedlings. It means when the concentration of plant extract increased, the plant part growth decreased. On the other hand, the growth of root and shoot elongation showed a positive relationship with time, i.e., when the days increased, the plant part elongation (growth) was also found to increase in extract used of all concentrations.

The growth of roots and shoots in seedlings by the use of DCM and methanol of both plant parts was found higher than the control condition. At high concentrations (5 mg/mL), the methanol root extract of the *P. hysterophous* did not affect the elongation of shoots or roots.

A similar negative effect was found between the shoot and root elongation of the wheat seedling and the PCA biplot on the concentration of DCM and methanol leaf extract (Figure 8) and root extract (Figure 9) of the *Xanthium strumarium*. In other words, the growth of root and shoot elongation showed a positive relationship with time, meaning that as the number of days increased, the plant part elongation (growth) also increased in the extract used of all concentrations. This indicates that the stem and root elongation decreased as the concentration of plant extract increased.

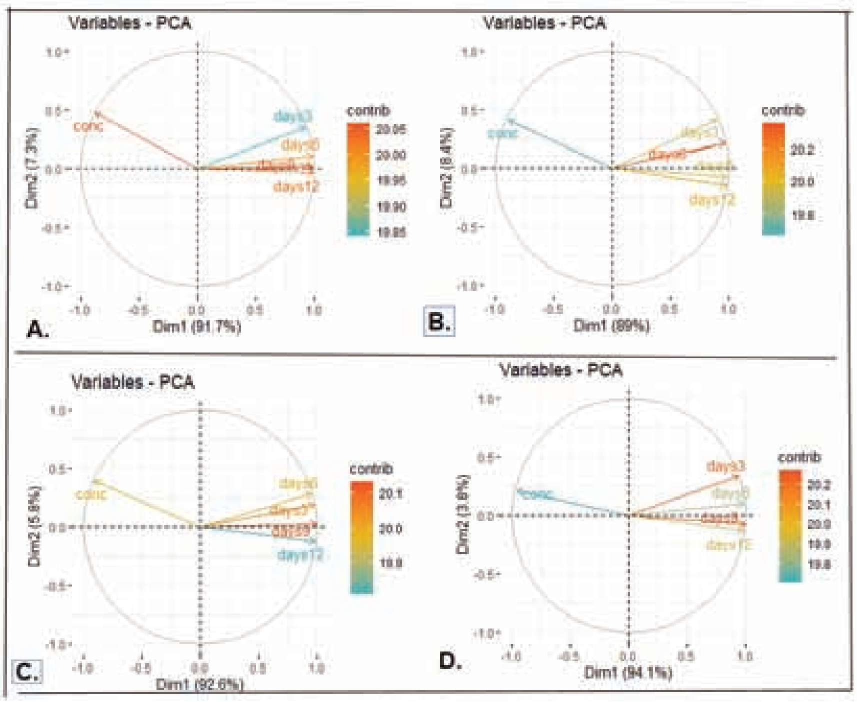


Figure 7: Bi-plot of individuals and variables on the effect of root extract on DCM (**A.** root growth & **B.** shoot growth) and on Methanol (**C.** root growth & **D.** shoot growth) of *P. hysterophorus* on wheat seed germination

At high concentrations (5 mg/mL), the DCM root extract of the *Xanthium strumarium* did not affect the elongation of shoots or roots. In most cases, the growth rate of the root and shoot of the treated plant was found higher than the high concentration and controlled condition. A similar result was found in the germination and growth of turnip, spinach, and ladies' finger by treatment of aqueous extract of *Terminalia belerica* in comparison with control (Talukder et al., 2015).

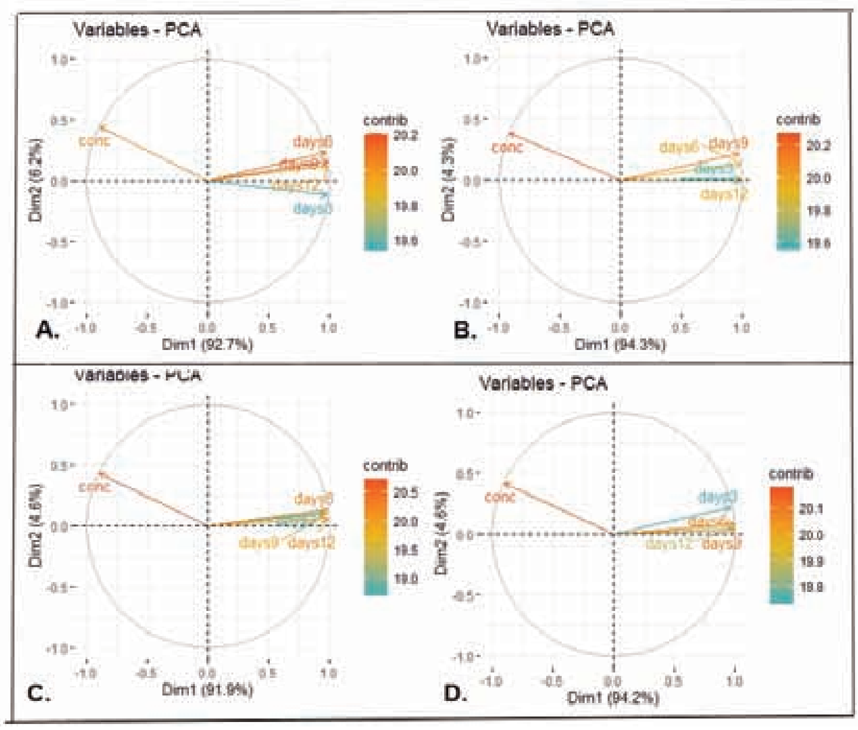


Figure 8: Bi-plot of individuals and variables on the effect of leaf extract on DCM (**A.** root growth & **B.** shoot growth) and on Methanol (**C.** root growth & **D.** shoot growth) of *Xanthium strumarium* on wheat seed germination

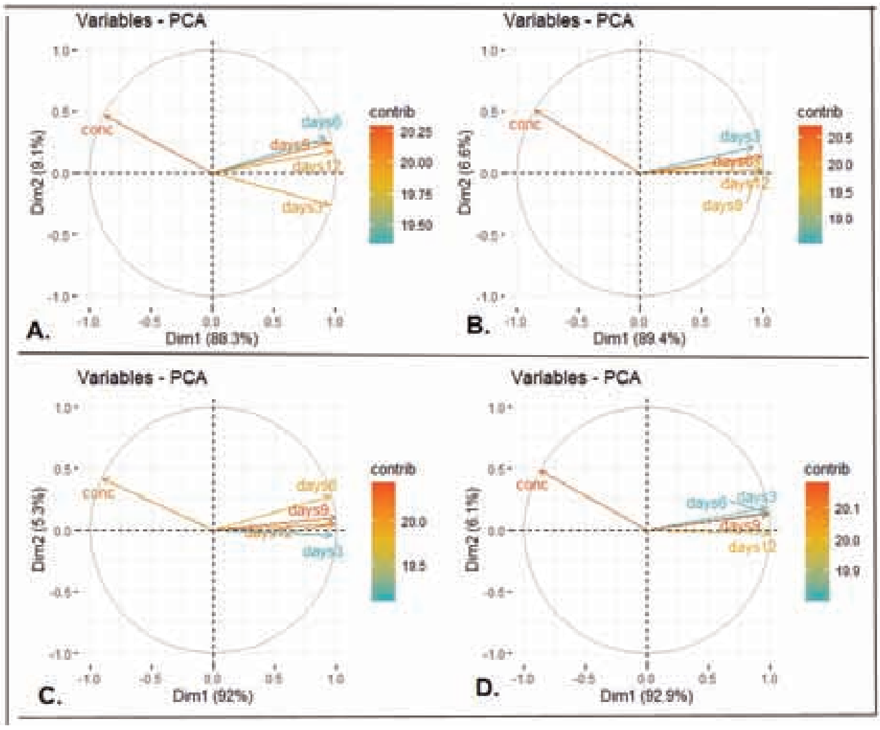


Figure 9: Bi-plot of individuals and variables on the effect of root extract on DCM (**A.** root growth & **B.** shoot growth) and on Methanol (**C.** root growth & **D.** shoot growth) of *Xanthium strumarium* on wheat seed germination

Assessment of brine shrimp lethality assay using various extracts of P. hysterophorus and X. strumarium

This study found that DCM and methanol extracts of *X. strumarium* leaf and root extracts showed higher cytotoxicity to brine shrimp nauplii than *P. hysterophorus* leaf and root extracts. The lethal concentration 50% (LC_{50}) is a widely utilized measurement to determine the cytotoxicity of substances or extracts, especially in toxicological and pharmaceutical studies. A lower LC_{50} value for extracts indicates higher cytotoxicity. The root DCM extract of *X. strumarium* had the highest cytotoxic activity (LC_{50}: 129.16 µg/mL), followed by the root methanol extract of *X. strumarium*, the root DCM extract of *P. hysterophorus*, and the leaf methanol extract of *P. hysterophorus* against brine shrimp nauplii (Table 7). The percentage mortality of brine shrimp nauplii was dosage-dependent after 24 hours of incubation.

The cytotoxicity of extracts was compared with a positive control, potassium dichromate (LC_{50} = 65.5 µg/mL). Meyer et al. (1982) defined cytotoxicity as a chemical compound or extract with an LC_{50} value of less than 1000 µg/mL in the brine shrimp lethality test (BSLA). According to Oketch-Rabah

Table 7: LC_{50} values of various extracts of *P. hysterophorus* and *X. strumarium*

Plants	Plant parts	Extracts	IC_{50} (µg/mL)
Parthenium hysterophorus	Leaf	DCM	438.24
		Methanol	497.44
	Root	DCM	302.93
		Methanol	395.48
Xanthium strumarium	Leaf	DCM	293.03
		Methanol	392.31
	Root	DCM	129.16
		Methanol	216.38
Potassium dichromate (positive control)	IC_{50}: 65.5 µg/mL		

et al. (1999) the indicated cytotoxic activity is poor at LC_{50} of 500-1000 μg/mL, medium at 100-500 μg/mL, and strong at 0-100 μg/mL. Therefore, all the extracts of *P. hysterophorus* and *X. strumarium* were moderately cytotoxic to brine shrimp nauplii. However, the root extracts of both plants were more cytotoxic than the leaf extracts, and the DCM extracts of both plants were more cytotoxic than the methanol extracts on the brine shrimp nauplii. It shows that the concentration of allelochemicals depends on the plant parts. Earlier studies found that DCM extract was more cytotoxic to nauplii than other types of extracts from other plants, including *Lilium nepalense* D. Don (Thapa et al., 2023), *Tephrosia purpurea* (L.) Pers., *Andrographis paniculata* (Burm.f.) Wall. ex Nees, and *Oldenlandia umbellata* L. (Suneka and Manoranjan, 2021).

Relationship between the phytotoxicity and cytotoxicity of various extracts of P. hysterophorus and X. strumariumto wheat seedlings and brine shrimp nauplii

According to this study, the specific extracts with allelochemicals of *P. hysterophorus* and *X. strumarium* that exhibited higher phytotoxicity to wheat seedlings (plant cells) also typically had higher cytotoxicity to brine shrimp nauplii (animal cells). It might be because allelochemicals in plants and animals work similarly. For instance, the DCM extracts of *X. strumarium* leaves and roots showed more cytotoxicity to brine shrimp nauplii and greater inhibition to the growth of roots and shoots in wheat seedlings than those of methanol extracts of *X. strumarium* leaves and roots. Similarly, in the case of plant parts, the roots of both plants showed higher cytotoxicity to brine shrimp nauplii and greater inhibition to the growth of roots and shoots in wheat seedlings than the leaves. This study can infer that the extract contains allelochemicals or phytotoxins that have growth-inhibitory or general cytotoxic effects. These results indicate that the extract might have a non-specific mechanism affecting both plants and animals, and broad-spectrum biological action, which could include antibacterial, anticancer, or herbicidal effects (Weston & Duke, 2003). According to Mousavi et al. (2021) allelopathic and cytotoxic properties are often linked with phenolic compounds (such as caffeic acid, rosmarinic acid) and terpenoids (such as carvacrol, thymol).These substances impair cellular processes, such as the integrity of membranes, mitochondrial activity, and oxidative balance, in aquatic organisms and plants (Pinheiro et al., 2015). *Juglans regia* releases allelochemicals that reduce nearby weeds and exhibit cytotoxicity to brine shrimp, indicating broader antibacterial or antipredator properties (Dorąevic et al., 2022). However, methanol extracts of leaf and roots of *P. hysterophorus* showed higher inhibition to the growth of roots and shoots in wheat seedlings, and the DCM extracts of leaf and roots showed higher cytotoxicity to brine shrimp nauplii.

Conclusion

This study showed that the allelochemicals or phytotoxins found in the methanol and DCM extracts of the roots and leaves of *P. hysterophorus* and *X. strumarium* were phytotoxic to wheat seeds and seedlings, and cytotoxic to brine shrimp nauplii. The concentration of allelochemicals may vary across various types of solvent extracts, and their distribution may vary among plant parts. The root methanol extract of *P. hysterophorus* was more effective at inhibiting seed germination and the growth of roots and shoots in wheat seedlings; however, the root DCM extract of *X. strumarium* was more effective in inhibiting the growth of roots and shoots in wheat seedlings, indicating that the nature (polar or non-polar) of phytotoxic chemicals differ based on plant species. Similarly, root DCM extracts of *X. strumarium* and *P. hysterophorus* were more toxic to brine shrimp nauplii than leaf methanol extracts. Moreover, the roots of both plants were more phytotoxic to seedling growth and cytotoxic to brine shrimp nauplii than the leaves, indicating that the roots contained a higher concentration of allelochemicals. This study also exhibits that allelochemicals present in both plant extracts have similar modes of action for phytotoxicity to seedling growth and cytotoxicity to brine shrimp nauplii. This study will be helpful for future research into the effect of allelochemicals on natural vegetation and crop plants, as well as the anticancer activity of allelochemicals found in *P. hysterophorus* and *X. strumarium*on various human cell lines.

Author Contributions

C B Thapa did conceptualization, methodology, formal data analysis, writing draft, and final editing and U Thapa did plant collection, data curation, and lab work. B R Nepali did Data analysis, and editing.

Acknowledgements

We would like to thank the Department of Botany, Butwal Multiple Campus for providing lab facilities for this research.

References

Adkins, S. W. & Shabbir, A. (2014). Biology, ecology, and management of the invasive parthenium weed (*Parthenium hysterophorus* L.). *Pest Management Science*, *70*, 1023-1029.

Amare, T. (2018). Allelopathic effect of aqueous extracts of parthenium (*Parthenium Hysterophorus* L.) parts on seed germination and seedling growth of maize (*Zea Mays* L.). *Journal of Agriculture and Crops*, *4*(12), 157-163. https://doi.org/10.32861/jac.412.157.163

Banik, D., & Yomso, J. (2021). *Hand book of weed identification*. Educreation Publication.

Bashar, H. M. K., Juraimi, A. S., Ahmad-Hamdani, M. S., Uddin, M. K., Asib, N., Anwar, M. P., Rahaman, F., Haque, M. A., & Hossain, A.(2023). Evaluation of allelopathic effects of *Parthenium hysterophorus* L. methanolic extracts on some selected plants and weeds. *PLoSONE*, *18*(1), e0280159. https://doi.org/10.1371/journal.pone.0280159

Bashar, H. M. K., Juraimi, A. S., Ahmad-Hamdani, M. S., Uddin, M. K., Asib, N., Anwar, M. P., & Rahaman, F. (2021). A mystic weed, *Parthenium hysterophorus*: Threats, potentials and management. *Agronomy*, *11*(8), 1514. https://doi.org/10.3390/agronomy11081514

Benyas, E., Hassanpouraghdam, M. B., Salmasi, S. Z., & Oskooei, O. S. K. (2010). Allelopathic effects of *Xanthium strumarium* L. shoot aqueous extract on germination, seedling growth and chlorophyll content of lentil (*Lens culinaris* Medic.). *Romanian Biotechnological Letters*, *15*(3), 5223-5228.

Bewley, J. D., Bradford, K. J., Hilhorst, H. W. M., & Nonogaki, H. (2013). *Seeds: Physiology of development, germination and dormancy* (3rd ed). Springer. https://doi.org/10.1007/978-1-4614-4693-4_3

Calabrese, E. J., & Baldwin, L. A. (2003). Hormesis: The dose-response revolution. *Annual Review of Pharmacology and Toxicology*, *43*(1), 175-197. https://doi.org/10.1146/annurev.pharmtox.43

Cheng, F., & Cheng, Z. (2015). Research progress on the use of plant allelopathy in agriculture and the physiological and ecological mechanisms of allelopathy. *Frontiers in Plant Science*, *6*, 1020. https://doi.org/10.3389/fpls.2015.01020.

Chinnuswamy, P., Chandran, K., & Mahadevaiah, M. (2018). *Tropical Weeds: Biology & identification*. Narendra Publishing House.

Chou, C. H. (1999). Roles of allelopathy in plant biodiversity and sustainable agriculture. *Critical Reviews in Plant Sciences*, *18*(5), 609-636.

Dorąevic, T., DurovicPejcev, R., Stevanovic, M., SaricKrsmanovic, M., Radivojevic, Lj., Santric, Lj., & GajicUmiljendic, J. (2022). Phytotoxicityand allelopathic potentialof *Juglans regia* L. leaf extract. *Front. Plant Sci.*, *13*, 986740. https://doi.org/10.3389/fpls.2022.986740

Duke, S. O., & Lydon, J. (1987). Herbicides from Natural Compounds. *Weed Technology*, *1*(2), 122-128. http://www.jstor.org/stable/3986711

Dukpa, R., Tiwari, A., & Kapoor, D. (2020). Biological management of allelopathic plant *Parthenium* sp. *Open Agriculture*, *5*(1), 252-261. https://doi.org/10.1515/opag-2020-0027

Einhellig, F. A. (1994). Mechanism of action of allelochemicals in Inderjit. In K. M. M. Dakshini, & Frank A. Einhellig (Eds), *Allelopathy, (Organisms, Processes, and Applications), Mechanism of Action of Allelochemicals in Allelopathy, ACS Symposium* Series, (Vol. 582) (pp. 96-116). American Chemical Society**.** https://doi.org/10.1021/bk-1995-0582.ch007

El-Gawad, A. A., Elshamy, A., El Gendy, A. E.-N., Gaara, A., & Assaeed, A. (2019). Volatiles profiling, allelopathic activity, and antioxidant potentiality of *Xanthium Strumarium* leaves essential oil from Egypt: Evidence from chemometrics analysis. *Molecules*, *24*(3), 584. https://doi.org/10.3390/molecules24030584

Fatope, M. O., Ibrahim, H., & Takeda, Y. (1993). Screening of higher plants reputed as pesticides using the brine shrimp lethality assay. *International Journal of Pharmacognosy, 31*, 250-256.

Gniazdowska, A., & Bogatek, R. (2005). Allelopathic interactions between plants. Multi-site action of allelochemicals. *Acta Physiologiae Plantarum, 27*(3), 395-407. https://doi.org/10.1007/s11738-005-0017-3

Harborne, J. B. (1998). *Insecticides of natural origin*. *Phytochemistry,* 48(3), 409-413.

He, H. B., Wang, H., Fang, C. X., Lin, Z. H., Yu, Z. M., & Lin, W. X. (2012). The role of volatile allelochemicals of *Eucalyptus* in interspecific interaction and allelopathy. *Journal of Chemical Ecology, 38*(4), 498-508. https://doi.org/10.1007/s10886-012-0106-9

Hussain, Z., Marwat, K. B., Cardina, J., & Khan, I. A. (2014). *Xanthium strumarium* L. impact on corn yield and yield components. *Turkish Journal of Agriculture and Forestry,38*(1), 39-46.

Inderjit, & Duke, S. O. (2003). Ecophysiological aspects of allelopathy. *Planta*, *217*(4), 529-539. https://doi.org/10.1007/s00425-003-1054-z

Invasive Species Specialist Group. (n.d.). *Global Invasive Species Database (GISD)*. IUCN. Retrieved June 29, 2025, from, https://www.iucngisd.org/gisd/about.php

Jalali, M., Moosavinasab, M., & Saffari, M. (2013). Allelopathic effects of aqueous extracts of *Xanthium strumarium* L. on germination characteristics and seedling growth of *Zea maize* L. *International Journal of Agriculture: Research and Review*, *3*(2), 223-227.

Kanchan, S. D., & Jayachandra. (1980). Allelopathic effects of *Parthenium hysterophorus* L. part IV. Identification of Inhibitors. *Plant and Soil, 55*(1), 67-75. https://doi.org/10.1007/bf02149710

Khan, M. A., Gulzar, S., & Hameed, A. (2011). Seed germination ecology of halophytes under saline conditions. *Environmental and Experimental Botany, 70*(2-3), 174-182.

Khan, N., Hashmatullah, Naveed, K., Zahid Hussain, Z., & Khan, S. A., (2012). Assessment of allelopathic effects of Parthenium (*Parthenium hysterophorus* L.) plant parts on seed germination and seedling growth of wheat (*Triticumaestivum* L.) cultivars. *Pak. J. Weed Sci. Res., 18*(1), 39-50.

Kroemer, G., Galluzzi, L., & Brenner, C. (2009). Mitochondrial membrane permeabilization in cell death. *Physiological Reviews, 87*(1), 99-163. https://doi.org/10.1152/physrev.00013.2006

Li, Z.-H., Wang, Q., Ruan, X., Pan, C.-D., & Jiang, D.-A. (2010). Phenolics and plant allelopathy. *Molecules*, *15*(12), 8933-8952. https://doi.org/10.3390/molecules15128933

Macías, F. A., Molinillo, J. M., Varela, R. M., & Galindo, J. C. (2007). Allelopathy: A natural alternative for weed control. *Pest Management Science, 63*(4), 327-348. https://doi.org/ 10.1002/ps.1342

Meyer, B. N., Ferrigni, N. R., Putnam, J. E., Jacobsen, L. B., Nichols, D. E., & McLaughlin, J. L. (1982). Brine shrimp: A convenient general bioassay for active plant constituents. *Planta Medica*, *45*(5), 31-34. https://doi.org/10.1055/s-2007-971236

Mirzaee, M., & Saeedipour, S. (2021). Allelopathic effects of *Xanthium strumarium* L. on germination and seedling growth of Mung bean (*Vigna radiata* L. Wilczek). *Iranian Journal Pulses Research*, *12*(2), 26-33. https://doi.org/10.22067/ijpr.v12i2.86650

Mousavi, S. S., Karami, A., Haghighi, T. M., Alizadeh, S., & Maggi, F. (2021). Phytotoxic potential and phenolic profile of extracts from *Scrophularia striata*. *Plants (Basel)*, *10*(1), 135. https://doi.org/10.3390/plants10010135

Naderi, R., Ali, K., Rehman, A., Rasmann, S., & Weyl, P. (2024). Estimating the impact on maize production by the weed *Parthenium hysterophorus* in Pakistan. *CABI Agric Biosci*, *5*(14), 1-5. https://doi.org/10.1186/s43170-024-00217-2

Oketch-rabah, H. A., Dossaji, J. F., & Mberu, E. K. (1999). Antimalarial activity of some Kenyan medicinal plants. *Pharm Biol*., *37*, 329-34.

Pinheiro, P. F., Costa, A.V., Alves Tde, A., Galter, I. N., Pinheiro, C. A., Pereira, A. F., Oliveira, C. M., & Fontes, M. M. (2015). Phytotoxicity and cytotoxicity of essential oil from leaves of *Plectranthus amboinicus*, carvacrol, and thymol in Plant Bioassays. *Journal of Agricultural and Food Chemistry 63*(41), 8981-8990. https://doi.org/10.1021/acs.jafc.5b03049

Rajbhandari, K. R., Thapa Magar, M. S., Kandel, D. R., & Khanal, C. (2016). *Plant Resources of Kailali, West Nepal*. District Plant Resource Office, Kailali, Nepal.

REAL CCS, (2014). Methods for testing plant response to composted material and its contamination by weed seeds and propagules. Method Code: OFW004-006, Version: 3.1.

Rice, E. L. (1984). *Allelopathy* (2nd ed) (pp. 422). Academic Press.

Seifu, A., Lulekal, E., Demissew, S., & Woldu, Z. (2024). Allelopathic potential of invasive alien plant species, *Xanthium strumarium* L., water and methanol extracts on germination and seedling growth of *Guizotia abyssinica* (L.f.) Cass., and *Linum usitatissimum* L. *Journal of Biological Studies*, *7*(3), 70-94, https://doi.org/10.62400/jbs.v7i3.10605

Shajie, E. & Saffari, M. (2007). Allelopathic effects of cocklebur (*Xanthium strumarium* L.) on germination and seedling growth of some crops. *Allelopathy Journal*, *19*(2), 501-506.

Shakya, B., Budhathoki, S., Maharjan, S. R., & Thapa, L. B. (2021-22). Effects of invasive *Parthenium hysterophorus* leachates on seed germination and seedling growth of wheat (*Triticum aestivum*). *Journal of Natural History Museum*, *32*, 65-76.

Singh, H. P., Batish, D. R., & Kohli, R. K. (2003). Allelopathic interactions and allelochemicals: New possibilities for sustainable weed management. *Critical Reviews in Plant Sciences*, *22*(3-4), 239-311. https://doi.org/10.1080/713610858

Singh, H. P., Batish, D. R., Kohli, R. K., Saxena, D. B., & Arora, V. (2002). Effect of parthenin-A sesquiterpene lactone from *Parthenium Hysterophorus*-on early growth and physiology of *Ageratum Conyzoides* Harminder. *Journal of Chemical Ecology*, *28*(11), 2169-2179. https://doi.org/10.1023/a:1021089013754.

Suneka, & Manoranjan (2021). Brine shrimp lethality assay with selected medicinal plant extracts. *Vingnanam Journal of Science*, *16*(2), 14-17.

Talukder, M. A., Rahaman, M., Roy, B. & Saha, K. C. (2015). Effects of herbal plant extracts on germination and seedling growth of some vegetables. *International Journal of Science and Nature*, *6* (3), 421-425.

Thapa, C. B., Bhattarai, A., Pant, K. K., Bhattarai, H. D., & Pant, B. (2023). Evaluation of antioxidant, antidiabetic, and cytotoxic activities of *Lilium nepalense* D. DON. *Journal of Institute of Science and Technology*, *28*(2), 63-70. https://doi.org/10.3126/jist.v28i2.61174.

Waheed, M., Haq, S. M., Arshad, F., Vitasović-Kosić, I., Bussmann, R. W., Hashem, A., & Abd-Allah, E. F. (2024). *Xanthium strumarium* L., an invasive species in the subtropics: Prediction of potential distribution areas and climate adaptability in Pakistan. *BMC Ecology and Evolution*, *24*, 124. https://doi.org/10.1186/s12862-024-02310-6

Wang, H., Khor, T. O., Shu, L., Su, Z. Y., Fuentes, F., Lee, J. H., & Kong, A. N. (2012). Plants vs. cancer: A review on natural phytochemicals in preventing and treating cancers and their druggability. *Anticancer Agents Med Chem.*, *12*(10), 1281-1305. https://doi.org/10.2174/1871520128038330026

Weston, L. A., & Duke, S. O. (2003). Weed and crop allelopathy. *Critical Reviews in Plant Sciences*, *22*, 367-389. https://doi.org/10.1080/713610861

Zahid, H., Marwat, K. B., Cardina, J., & Khan, I. A. (2014). *Xanthium strumarium* L. impact on corn yield and yield components, *Turkish Journal of Agriculture and Forestry*, *38*(1), 39-46. https://doi.org/10.3906/tar-1210-53

Appendix 1: Result of individuals based on the explained variance of PCA analysis on the effect of DCM and methanol extracts of *Parthenium hysterophorus* on the growth of wheat seedlings

Used plant parts	**Particular**	**DCM**				**Methanol**			
		Root length		**Root length**		**Root length**		**Root length**	
		PCA1	PCA1	PCA1	PCA1	PCA1	PCA2	PCA1	PCA2
Leaf extract	Standard deviation	2.14	2.14	2.14	2.14	2.14	0.48	2.17	0.48
	Proportion of variance	0.91	0.92	0.92	0.92	0.92	0.05	0.94	0.05
	Cumulative proportion	0.91	0.92	0.92	0.92	0.92	0.96	0.94	0.98
Root extract	Standard deviation	2.14	2.15	2.15	2.15	2.15	0.51	2.17	0.55
	Proportion of variance	0.92	0.93	0.93	0.93	0.93	0.05	0.93	0.06
	Cumulative proportion	0.92	0.93	0.93	0.93	0.93	0.97	0.34	0.96

Appendix 2: Result of individuals based on the explained variance of PCA analysis on the effect of DCM and methanol extracts of *Xanthium strumarium*on the growth of wheat seedlings

Used plant parts	**Particular**	**DCM**				**Methanol**			
		Root length		**Shoot length**		**Root length**		**Shoot length**	
		PCA1	PCA2	PCA1	PCA2	PCA1	PCA2	PCA1	PCA2
Leaf extract	Standard deviation	2.15	0.55	2.17	0.46	2.14	0.48	2.17	0.48
	Proportion of variance	0.93	0.06	0.93	0.04	0.92	0.05	0.94	0.05
	Cumulative proportion	0.93	0.98	0.93	0.98	0.92	0.96	0.94	0.98
Root extract	Standard deviation	2.10	0.67	2.11	0.57	2.14	0.51	2.17	0.55
	Proportion of variance	0.88	0.09	0.89	0.07	0.92	0.05	0.93	0.06
	Cumulative proportion	0.88	0.97	0.89	0.96	0.92	0.97	0.34	0.96

Journal of Plant Resources (2025) **23**(1), 31-44
https://doi.org/10.3126/bdpr.v23i1.83349

GC-MS Analysis of Essential Oils from Selected Aromatic Plants of the Manang Region, Nepal

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Abstract

This study investigated the chemical composition of essential oils extracted from seven aromatic plant species collected from Manang district, Nepal; *Artemisia dubia* Wall. ex Besser, *Elsholtzia fruticosa* (D. Don) Rehder, *Tetrataenium lallii* (C.Norman) Cauwet, Carb & M.Farille, *Juniperus indica* Bertol., *Nepeta ciliaris* Benth., *Origanum vulgare* L., and *Thymus linearis* Benth. Essential oils were obtained through hydro distillation using the Clevenger apparatus and analyzed using Gas Chromatography-Mass Spectrometry (GC-MS). The analysis revealed a diverse array of terpenoid compounds, predominantly monoterpenes, sesquiterpenes, phenols and esters, in the investigated plant species. *Artemisia* and *Elsholtzia* contained γ-terpinene and terpene derivatives as the main components. Chamazulene was also found in the *Artemisia* essential oil. *Heracleum* was characterized by esters (esters of acetic acid and butanoic acid) and D-limonene, while *Juniperus* contained D-limonene, sabinene, and elemol as major constituents. *Nepeta* was rich in bergamotene, caryophyllene oxide, and sesquisabinene. *Origanum* primarily consisted of p-cymene and thymol, and *Thymus* essential oil was highly rich in thymol and carvacol. This research contributes to expanding our understanding of the chemical diversity of essential oils from high-altitude aromatic plants in the Nepalese Himalayas, providing valuable information for potential applications in various industries. The identification of novel compounds and the understanding of their biological activities could lead to the development of new natural products with potential applications in pharmaceuticals, cosmetics, and food industries.

Keywords: *Nepeta ciliaris*, *Origanum vulgarae*, Secondary metabolites, Sesquiterpenes, Terpenes, *Tetrataenium lallii, Thymus linearis*

Introduction

Essential oils are high-value, low-volume commodities primarily composed of complex mixtures of volatile, low-molecular-weight terpenoid and phenylpropanoid compounds (Mohamed & Alotaibi, 2022). These unique chemical constituents are specific to each plant, and are responsible for the oils' distinctive fragrances (Globe Newswire, 2022). Essential oils are extracted from aromatic plants. The extracted oils find diverse applications in cosmetics (Guzmán & Lucia, 2021), flavours and fragrances (Jugreet et al., 2021), spices (Cardoso-Ugarte & Sosa-Morales, 2021), repellents (Nerio et al., 2010), pesticides (Garrido-Miranda et al., 2022), herbal beverages (Christopoulou et al., 2021), aromatherapy (Ali et al., 2015), and even in radioprotection (Samarth et al., 2017).

Citrus oils, particularly lemon and orange essential oils, dominate the global market due to their exceptional functional and sensory properties. These oils are rich in a variety of compounds, including α-, β-pinene, sabinene, β-myrcene, *D*-limonene, linalool, α-humulene, and α-terpineol. These compounds belong to different chemical groups such as monoterpenes, monoterpene aldehydes/alcohols, and sesquiterpenes, and are known for their antioxidant, antimicrobial, anticancer, and anti-inflammatory properties (Bora et al., 2020). Furthermore, the European Union has legally recognized several components of plant-based volatile oils, such as carvacrol, carvone, cinnamaldehyde, citral, and p-cymene, as approved flavoring agents (*Essential Oils Market Size, Share & Growth Report 2021-2028*, n.d.).

Nepal's medicinal and aromatic plants (MAPs) sector, particularly its essential oils, is experiencing significant commercial growth. MAPs, encompassing a wide range of essential oil-bearing plants found both in natural and cultivated environments, constitute a crucial segment of non-timber forest products. These resources hold immense socio-cultural and economic value within Nepal with over 20 major essential oils demonstrating high export potential. While Nepal's initial essential oil exports were modest, they experienced remarkable growth between 2010 and 2021, surging at a compound annual growth rate (CAGR) of 11% from $974 to $7.76 million USD. This impressive trajectory established Nepal as the 52nd largest exporter of essential oils worldwide (Observatory of Economic Complexity [OEC], n.d.). Furthermore, the global essential oil market is poised for significant expansion. Valued at $21.79 billion in 2022, it is projected to grow at a CAGR of 7.9% from 2023 to 2030. This robust growth is primarily fueled by the increasing demand for essential oils across diverse sectors, including aromatherapy, the food and beverage industry, and the beauty and personal care market (*Essential Oils Market Size, Share & Trends Analysis Report By Product (Orange, Cornmint, Eucalyptus), By Application (Medical, Food & Beverages, Spa & Relaxation), By Sales Channel, By Region, And Segment Forecasts, 2023 – 2030*, n.d.; Poudel, 2022).

The global market has shown considerable interest in Nepali essential oils across three key sectors. In particular, they have garnered significant interest as premium flavours (natural health and organic foods such as timur, curcuma, ginger, cinnamon, and wintergreen), high-end beauty and personal care products (rhododendron, ginger, jatamansi, butternut, palmarosa), and pharmaceutical applications as herbal traditional medicines (*Essential Oils Market Size, Share & Trends Analysis Report By Product (Orange, Cornmint, Eucalyptus), By Application (Medical, Food & Beverages, Spa & Relaxation), By Sales Channel, By Region, And Segment Forecasts, 2023 – 2030*, n.d.). In this context, a thorough understanding of the chemical constituents of the essential oils from unexplored plant species is also crucial. Knowledge of their chemical composition is essential for assessing their quality, identifying their biological properties, and determining their appropriate applications.

Building upon our previous research on the chemical constituents of essential oils from high-altitude aromatic plants in the Langtang region of Nepal (Pradhan et al., 2023), this study is trying to further investigate and identify the components of essential oils obtained from select aromatic plants in Manang district. Located in the Central Himalayan region, Manang district is renowned for its diverse medicinal plant species (Bhattarai et al., 2006). This region's unique high-altitude environment, climatic conditions and diverse flora are likely to yield essential oils with distinct chemical compositions and potential biological activities. For this study, we selected seven aromatic plant species, *Artemisia dubia* Wall. *ex* Besser, *Elsholtzia fruticosa* (D. Don) Rehder, *Tetrataenium lallii* (C.Norman) Cauwet, Carb & M. Farille (synonym: *Heracleum lallii)*, *Juniperus indica* Bertol., *Nepeta ciliaris* Benth., *Origanum vulgare* L. and *Thymus linearis* Benth.

This study employed advanced analytical techniques such as Gas Chromatography-Mass Spectrometry (GC-MS) to comprehensively analyse the volatile compounds present in the essential oils of the selected plant species. The findings of this research will contribute to a deeper understanding of the chemical diversity of essential oils from high-altitude plants in the Nepalese Himalayas. By expanding the knowledge on the chemical constituents of essential oils from this unique ecosystem, this study aims to contribute significantly to the field of natural products chemistry and support the sustainable development of the aromatic plants from Manang region, Nepal.

Materials and Methods

Plant collection

Aerial parts of seven wild plant species, *Artemisia dubia* Wall. *ex* Besser, *Elsholtzia fruticosa* (D. Don) Rehder, *Tetrataenium lallii* (C.Norman) Cauwet, Carb & M.Farille, *Juniperus indica* Bertol., *Nepeta ciliaris* Benth., *Origanum vulgare* L., and *Thymus*

Table 1: List of plant species collected from Manang District.

S.N.	Name	Family	Parts Used for Extraction	Location	Altitude (m)
1.	*Artemisia dubia* Wall. *ex* Besser	Asteraceae	Leaves	Pisang	3260
2.	*Elsholtzia fruticosa* (D. Don) Rehder	Lamiaceae	Leaves	Chame to Timang	2657
3.	*Tetrataenium lallii* (C.Norman) Cauwet, Carb & M.Farille	Apiaceae	Seeds	Tanki to Gunsang	3781
4.	*Juniperus indica* Bertol.	Cupressaceae	Leaves	Gunsang	3918
			Berries		
5.	*Nepeta ciliaris* Benth.	Lamiaceae	Leaves	Manang to Tanki	3700
			Flowers		
6.	*Origanum vulgare* L.	Lamiaceae	Aerial part	Gunsang	3918
7.	*Thymus linearis* Benth.	Lamiaceae	Aerial part	Gunsang	3906

linearis Benth., were collected from the Manang district, Nepal, in September 2022. The collection sites ranged from 2600 m to 3900 m in altitude. The collected plant species were shade-dried for two weeks. The collection of plant species, as listed in Table 1, was identified and confirmed by the National Herbarium and Plant Laboratories (KATH), Nepal.

Reagents

All chemicals used in this study were commercially available and used without further purification. Anhydrous sodium carbonate (analytical grade) was obtained from Fisher Scientific (Mumbai, India), while dichloromethane (HPLC grade) was sourced from Merck (Mumbai, India). Eugenol, *D*-limonene, *E*-caryophyllene, *p*-cymene, *α*-pinene, *α*-phellandrene, *β*-pinene, thymol, *δ*-3-carene, and eucalyptol were purchased from Sigma Aldrich (St. Louis, MO, USA).

Extraction of essential oil

Essential oil was extracted from shade-dried plant material using the hydro-distillation method (Clevenger, 1928). Approximately 30-40 grams of plant parts were placed in a 500 mL round bottom flask and subjected to hydro-distillation using a Clevenger apparatus. The extraction process was conducted for 4 hours, and the collected oil was stored in a glass vial. Any residual moisture in the essential oil was removed using anhydrous sodium carbonate, and the oil was subsequently stored under refrigerated conditions at 4°C.

Gas Chromatography-Mass Spectrometry (GC-MS) analysis

Essential oils were analysed using a Shimadzu GC-MS-QP2010 Plus system available at the Instrument Section of the Department of Plant Resources. A capillary column (SH-RTX-5MS, 60 m × 0.32 mm × 0.25 µm) with a 5% diphenyl/95% dimethyl polysiloxane stationary phase was employed. GC analysis was performed under the following conditions: column oven temperature, 50ºC; injection temperature, 250ºC; ion source temperature, 250ºC; interface temperature, 200ºC; split injection mode with a split ratio of 80; Helium with a pressure of 53.8 kPa; total gas flow, 112.3 mL/min; column flow, 1.35 mL/min. The GC-MS system starts with an initial oven temperature of 50ºC for 1 min, then increases to 230ºC at a rate of 3ºC for 9 min. Mass spectral detection was carried out in electron ionization mode with a scan range of 40-350 m/z. The total analysis time for each sample was 70 m.

The chemical components of the essential oils were identified by comparing their mass spectral fragmentation patterns with those in the National Institute of Standards and Technology (NIST) 2017 library and the Mass spectra of Flavour and Fragrance of Natural and Synthetic Compounds (FFNSC) 4.0 library and also by comparing the retention times of the components with those of reference compounds. The percentage composition of each component in the essential oil (Area %) is reported as raw percentages based on the total ion chromatogram (TIC) without any standardization.

Results and Discussion

Essential oils were obtained through hydrodistillation of the aerial parts of seven plant species: *Artemisia dubia*, *Elsholtzia fruticosa*, *Tetrataenium lallii*, *Juniperus indica*, *Nepeta ciliaris*, *Origanum vulgare*, and *Thymus linearis*. Their chemical composition was subsequently determined using GC-MS analysis. Appendix 1 provides a detailed list of the identified chemical constituents present in the essential oils of these plant species and Figure 1 depicts the structure of some of the identified components. Similarly, Figure 2 to Figure 10 depict the total ion chromatograms (TICs) of each essential oil. While not all components were identified, the majority of those detected were monoterpenes and sesquiterpenes. Additionally, phenolic compounds and esters were also identified in the essential oils extracted from some of these plant species.

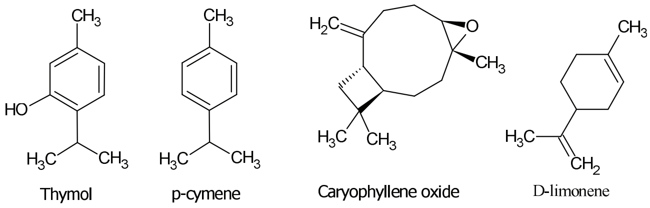

Figure 1: Structure of some of the identified terpenoid components

The essential oil extracted from the leaves of *Artemisia dubia* was dominated by terpene derivatives, including *γ*-terpinene, *α*-terpinene, *α*-terpinene geranyl, and terpinen-4-ol, collectively representing 23.8% of the oil. Minor constituents included chamazulene (6.7%), chrysanthenol (3.8%), caryophyllene derivatives (5.4%), nerolidol (3.3%), *p*-cymene (4.2%), naphthalene derivatives (8.4%), and keto derivatives such as and rostanone (2.5%). Previous studies on *Artemisia dubia* from the Langtang region at a similar altitude identified different major constituents, primarily comprising santolinatriene, *β*-cubebene, *β*-pinene, sabinene, *α*-pinene, *E*-caryophyllene, bicyclogermacrene, *α*-humulene, *E*-nerolidol, and lavandulyl acetate (Pradhan et al., 2023). These findings suggest that the chemical constituents of this plant species are influenced by climatic conditions (Alum, 2024). While both Langtang and Manang regions in Nepal are high-altitude regions with cold climates, they differ significantly in precipitation levels. Manang experiences significantly less rainfall due to the rain shadow effect, creating a considerably drier climate compared to Langtang, which receives more moisture. Essentially, Manang has a more desert-like climate compared to Langtang (Kharal et al., 2017; Steiner et al., 2021). Such differences in climatic conditions can significantly impact the vegetation in these areas and ultimately lead to variations in the chemical composition of the plant species. Similarly previous studies conducted by other researchers have also reported varying chemical compositions for the essential oil of *Artemisia dubia* from different locations. A study conducted in Kirtipur, Nepal, identified chrysanthenone, coumarin, and camphor as major components (Satyal et al., 2012). Similarly, Liang et al. (2017) reported terpinolene and limonene as major components in *Artemisia dubia* essential oil from China. These variations in chemical composition can likely be attributed to factors such as location, altitude, and the time of year plant material was collected. For instance, Satyal et al. (2012) collected their *Artemisia dubia* samples from hilly areas at an altitude of 1360 m in May, while this study utilized plant material collected in September from a mountainous region at a significantly higher altitude of approximately 3200 meters. These contrasting collection conditions likely contribute to the observed differences in the essential oil constituents.

The essential oil extracted from the leaves of Elsholtzia fruticosa primarily consisted of terpenoid compounds, including terpinen-4-ol (18.1%), *γ*-terpinene (16.5%), *α*-terpinene (8.0%), *α*-terpineol (3.8%), and *α*-terpineolene (3.8%). In addition to these terpene derivatives, other constituents such as *p*-cymene (9.6%), sabinene hydrate (4.6%), nerolidol (4.3%), and *α*-thujene (2.4%) were also identified. This chemical composition contrasts with previous studies on Elsholtzia fruticosa from the Langtang region (Pradhan et al., 2023), which, despite the similar altitude, revealed a different profile dominated by perillene, eucalyptol, *β*-pinene, and *E*-caryophyllene. This discrepancy further

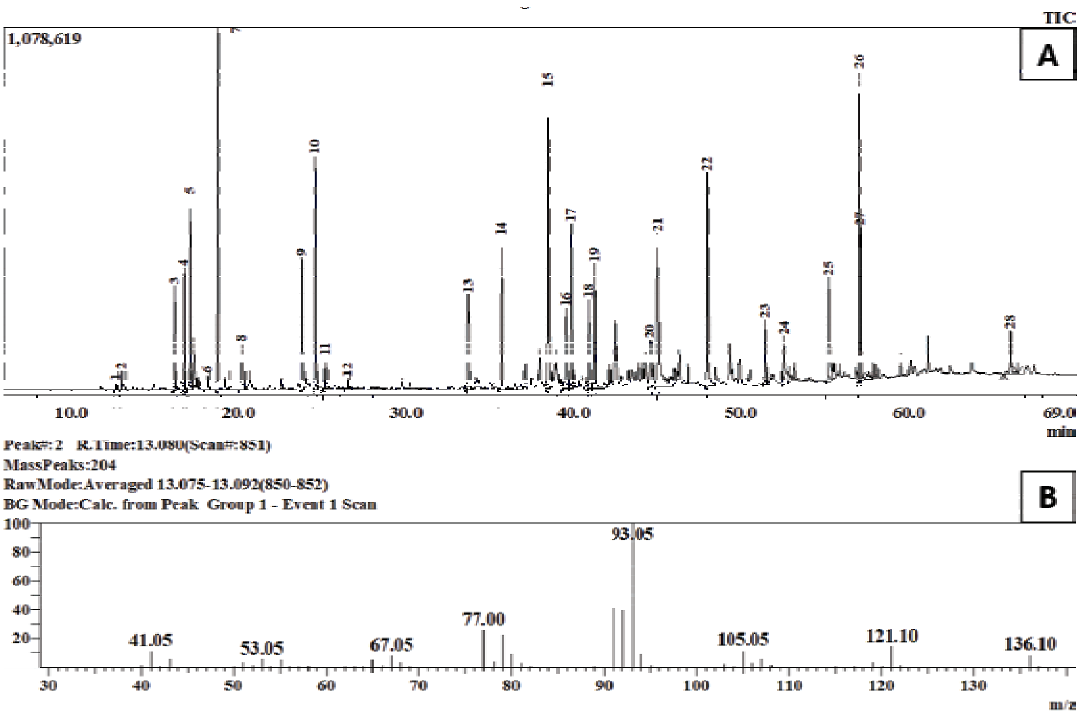

Figure 2: (A) TIC of essential oil from *Artemisia dubia*, **(B)** mass spectrum of peak #2 (α-pinene)

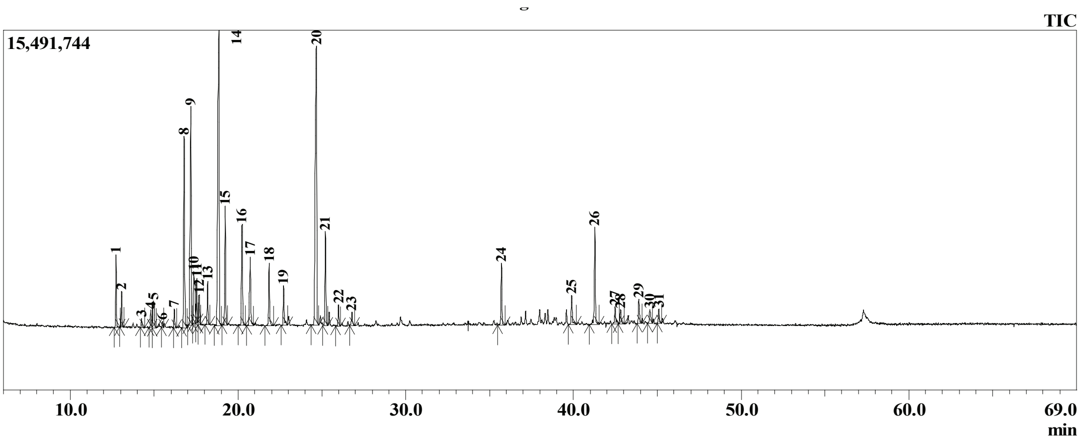

Figure 3: TIC of essential oil from *Elsholtzia fruticosa*

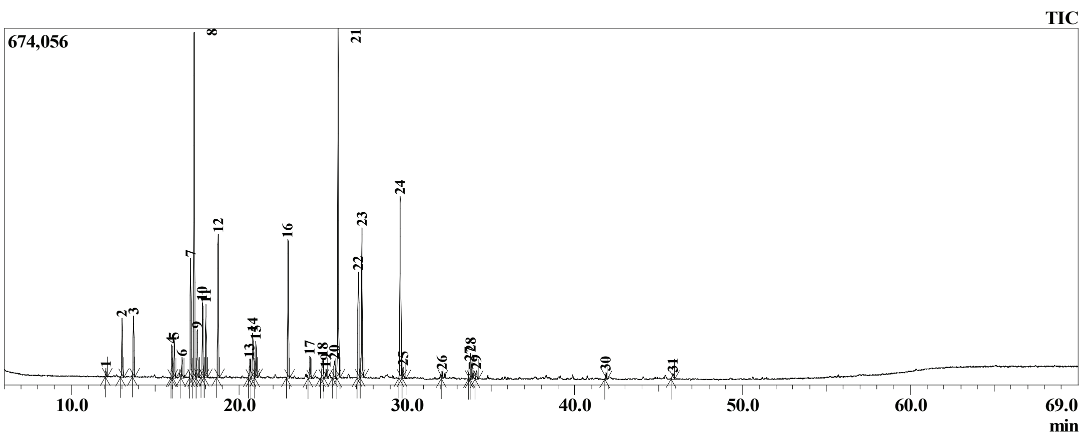

Figure 4: TIC of essential oil from *Tetrataenium lallii*

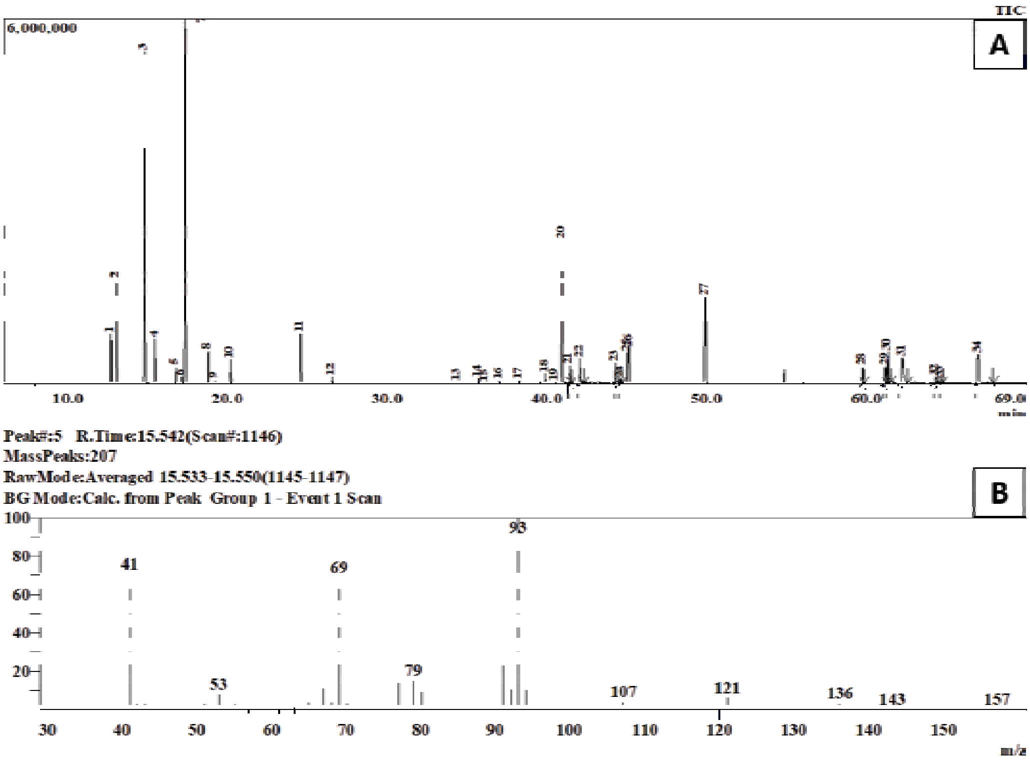

Figure 5: (A) TIC of essential oil from *Juniperus indica* berries, **(B)** mass spectrum of peak #5 (*β*-myrcene)

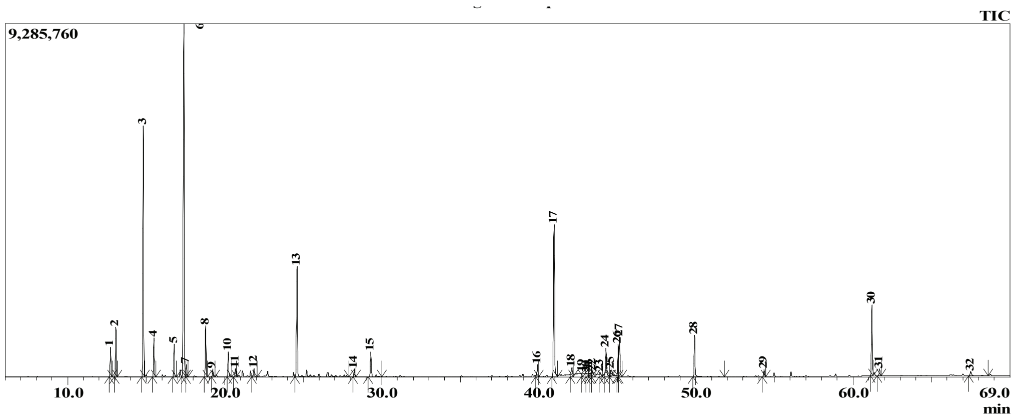

Figure 6: TIC of essential oil from *Juniperus indica* leaves

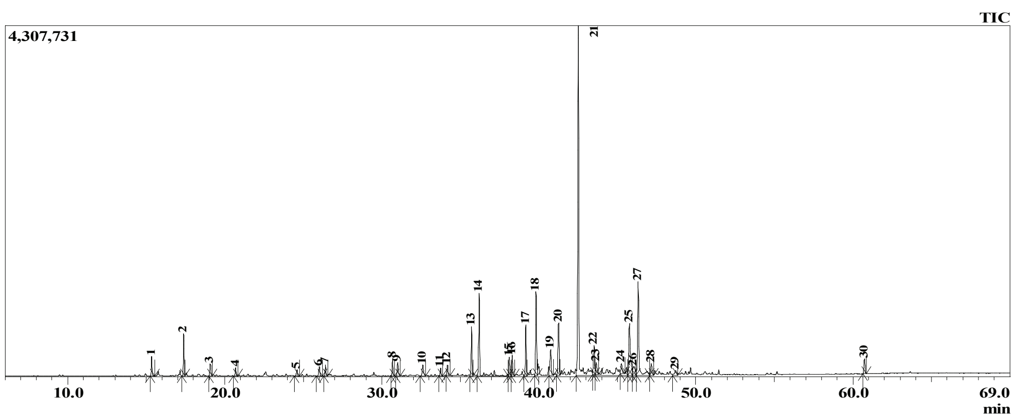

Figure 7: TIC of essential oil from *Nepeta ciliaris* (flowers)

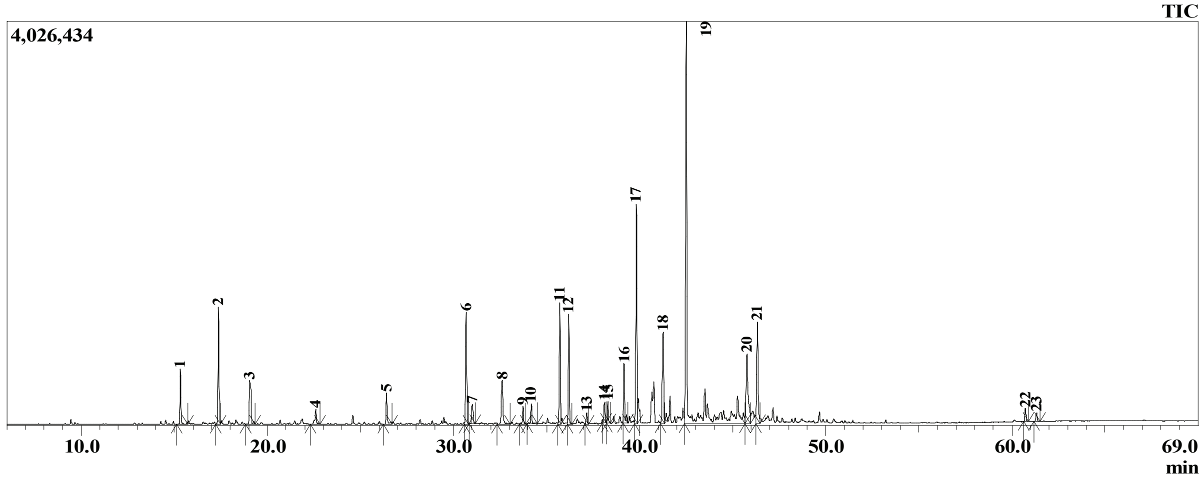

Figure 8: TIC of essential oil from *Nepeta ciliaris* (leaves)

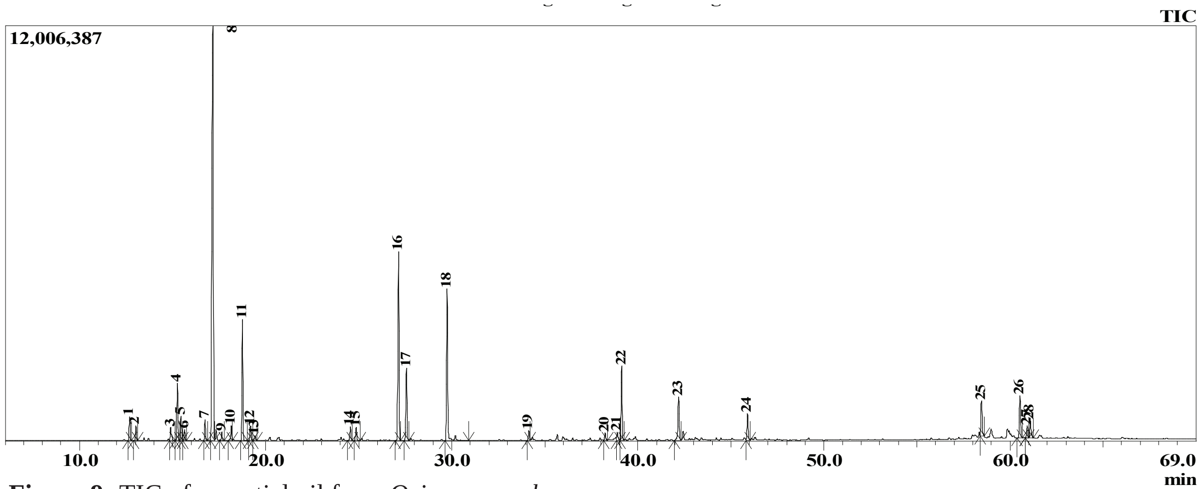

Figure 9: TIC of essential oil from *Origanum vulgare*

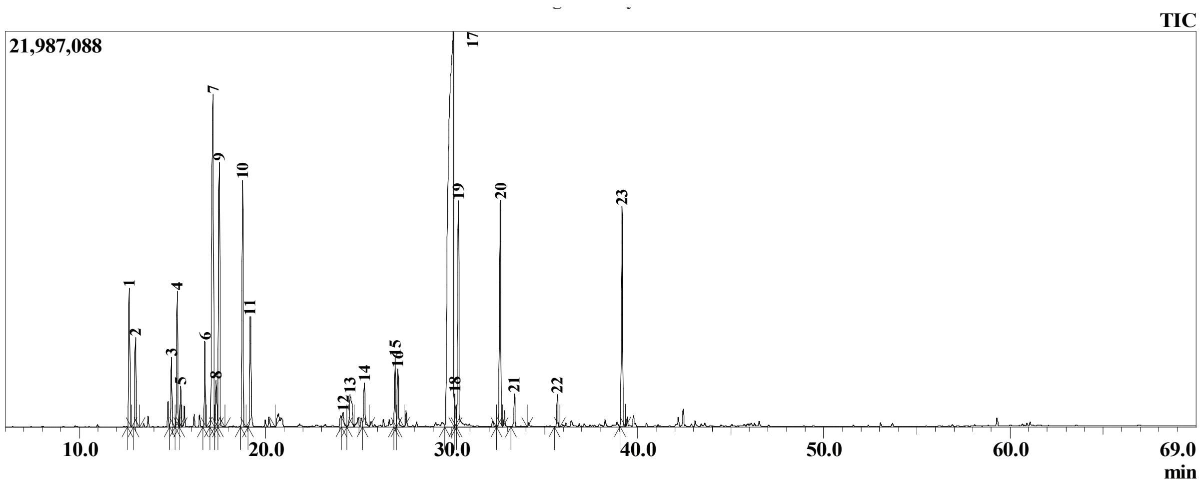

Figure 10: TIC of essential oil from *Thymus linearis*

emphasizes the significant influence of climatic variations on the chemical composition of plant species, even within the same region at similar altitudes. However, the constituents of the essential oil from the seeds of *Tetrataenium lallii* (synonym: *Heracleum lallii)* closely resembled those reported in the literature for other *Heracleum* species. Mustafavi et al. (2022) identified the essential oil from Iranian populations of *Heracleum persicum* as being highly rich in hexyl butyrate and octyl acetate. Similarly, Jagannath et al. (2012) identified bornyl acetate, *α*-pinene, limonene, and octyl acetate as the major constituents in the seeds of the Indian species, *Heracleum rigens*. While the specific constituents may vary between species, these studies consistently demonstrate that ester derivatives are major components of the essential oils extracted from the seeds of *Heracleum* species. Our findings from *T. lallii* support this observation. The seeds of *T. lallii* predominantly contained ester derivatives, including butanoic acid esters (15.9%), acetic acid octyl ester (15.7%), bornyl acetate (8.6%), *p*-cymene (5.3%), isobutyrates (6.5%), butyrates (6.1%), butyl isovalerate (3.1%), along with *D*-limonene (15.4%).

The essential oil extracted from the berries and leaves of *Juniperus indica* was rich in monoterpenoids, predominantly limonene and sabinene, and the sesquiterpenoid, *α*-elemol. Minor components included *α*-pinene, *α*-terpinene, *α*-thujene, myrcene, eudesmol, and cadinene. The chemical composition of the essential oils from berries and leaves exhibited some similarities. While both contained limonene, the leaf oil had a higher concentration (28.5%) compared to the berry oil (23.9%). Conversely, the berry oil was significantly richer in sabinene (20.2%) than the leaf oil (15.8%). Notably, *α*-elemol was present at approximately 10% in both cases. These findings differ from our previous studies on the essential oil of *Juniperus recurva* from the Langtang region, which primarily comprised *δ*-3-carene, *β*-pinene, cadina-1(6), 4-diene, *δ*-cadinene, and *α*-terpinolene. Another study conducted on the leaves and berries of *J. indica* from Uttarakhand at an altitude of 3800 meters revealed a somewhat similar profile, with sabinene, terpinen-4-ol, *α*-pinene, and *γ*-terpinene as the major components (Lohani et al., 2010). Essential oils extracted from *Nepeta* species have been primarily reported to be rich in caryophyllene, sesquiphellandrene, and caryophyllene oxide, along with trace amounts of other sesquiterpenoid compounds (Baranauskiene et al., 2019; Kumar & Mathela, 2018; Kumar et al., 2019). Similarly, the essential oils extracted from both the leaves and flowers of *Nepeta* were found to be predominantly composed of caryophyllene oxide (29.0% in leaves, 34.4% in flowers), sesquisabinene (10.8% in leaves, 5.8% in flowers), caryophyllene (6.0% in leaves, 3.5% in flowers), and bergamotene (6.2% in leaves, 7.2% in flowers), which aligns well with previous literature. In addition to these major sesquiterpenes, cyclohexenone derivatives were also identified as minor constituents (5.9% in leaves, 8.9% in flowers).

Oreganum vulgare and *Thymus linearis* are primarily used as culinary herbs. The essential oils extracted from the aerial parts of both plant species were rich in oxygenated compounds. *Oreganum vulgare* primarily consisted of *p*-cymene (34.1%), thymol methyl ether (12.1%), thymol (10.1%), terpinene (6.9%), bisabolene (4.5%), and carvacryl methyl ether (4.5%). *Thymus linearis* was notably rich in thymol (41.3%), alongside *p*-cymene (12.1%), thymol acetate (6.7%), carvacrol (6.2%), bisabolene (5.8%), eucalyptol (5.6%), and thujene (2.5%). Both plant species exhibited similar essential oil profiles, with variations in the relative abundance of carvacrol, *p*-cymene, thymol, and terpinene, along with other minor constituents (Firdous et al., 2023; Han et al., 2017; Kabdal et al., 2022; Teixeira et al., 2013).

Conclusion

This study provides valuable insights into the chemical composition of essential oils extracted from seven aromatic plant species collected from Manang district, Nepal. The GC-MS analysis revealed a diverse array of chemical constituents, with terpenoids, particularly monoterpenes and sesquiterpenes, being the predominant compounds. Key findings include the identification of compound, chamazulene, in *Artemisia dubia* and the characterization of the essential oil of *Nepeta ciliaris* essential oil, which was found to be primarily composed of sesquisabinene, caryophyllene oxide, and bergamotene. Furthermore, this study highlights the significant influence of factors such as altitude and climate on the chemical composition of essential oils. This is evidenced by comparing the composition of *Artemisis dubia* and *Elsholtzia fruticosa essential oils* with previous studies on similar species from different locations. These findings contribute to a deeper understanding of the chemical diversity of essential oils from high-altitude aromatic plants in the Nepalese Himalayas. This knowledge has potential applications in various fields, including pharmaceuticals, cosmetics, and the food industry. Further research is warranted to investigate the biological activities and potential therapeutic applications of these unique essential oil compositions.

Author Contributions

S Pradhan and D R Kandel collected the plant materials from Manang region; S Pradhan and R Maharjan dried the plant materials, extracted and analysed the essential oil; S Pradhan conducted literature survey and prepared the manuscript; D R Kandel prepared the herbarium and identified the collected plant materials; D R Kandel and R Maharjan reviewed the manuscript. All authors have read and agreed to the final version of the manuscript.

Acknowledgements

The authors express their sincere gratitude to Dr. Radha Wagle, former Director General, Mr. Saroj Kumar Chaudhary, Deputy Director General of the Department of Plant Resources, and Mr. Subhash Khatri, former Chief of the National Herbarium and Plant Laboratories (KATH), for their unwavering support. We also extend our heartfelt thanks to Dr. Madhu Sudan Thapa Magar and Dr. Seerjana Maharjan for their invaluable assistance with plant collection, herbarium preparation, and field guidance. Furthermore, the authors acknowledge the Department of Plant Resources for providing the opportunity to conduct this research and for their generous financial support.

References

Ali, B., Al-Wabel, N. A., Shams, S., Ahamad, A., Khan, S. A., & Anwar, F. (2015). Essential oils used in aromatherapy: A systematic review. *Asian Pacific Journal of Tropical Biomedicine*, *5*(8), 601-611.

Alum, E. U. (2024). Climate change and its impact on the bioactive compound profile of medicinal plants: Implications for global health. *Plant Signaling & Behavior, 19*(1).

Baranauskienė, R., Bendžiuvienė, V., Ragažinskienė, O., & Venskutonis, P. R. (2019). Essential oil composition of five *Nepeta* species cultivated in Lithuania and evaluation of their bioactivities, toxicity and antioxidant potential of hydrodistillation residues. *Food and Chemical Toxicology, 129*, 269-280.

Bhattarai, S., Chaudhary, R. P., & Taylor, R. S. L. (2006). Ethnomedicinal plants used by the people of Manang district, Central Nepal. *Journal of Ethnobiology and Ethnomedicine*, 2(1).

Bora, H., Kamle, M., Mahato, D. K., Tiwari, P., & Kumar, P. (2020). Citrus essential oils (CEOs) and their applications in food: An overview. *Plants, 9*(3), 357.

Cardoso-Ugarte, G. A., & Sosa-Morales, M. E. (2021). Essential oils from herbs and spices as natural antioxidants: Diversity of promising food applications in the past decade. *Food Reviews International, 38*(Sup1), 403-433.

Christopoulou, S. D., Androutsopoulou, C., Hahalis, P., Kotsalou, C., Vantarakis, A., & Lamari, F. N. (2021). Rosemary extract and essential oil as drink ingredients: An evaluation of their chemical composition, genotoxicity, antimicrobial, antiviral, and antioxidant properties. *Foods, 10*(12), 3143.

Clevenger, J. F. (1928). Apparatus for the determination of volatile oil. *The Journal of* *the American Pharmaceutical Association (1912)*, *17*(4), 345-349.

Essential Oils Market Size, Share & Growth Report, 2021-2028. (n.d.). https://www.fortunebusinessinsights.com/industry-reports/essential-oils-market-101063

Essential Oils Market Size, Share & Trends Analysis Report By Product (Orange, Cornmint, Eucalyptus), By Application (Medical, Food & Beverages, Spa & Relaxation), By Sales Channel, By Region, And Segment Forecasts, 2023 - 2030. (n.d.). https://www.grandviewresearch.com/industry-analysis/essential-oils-market#:~:text=Report%20Overview,7.9%25%20from%202023%20to%202030.

Firdous, S., Bhat, S. H., Aziz, S., M. Jehangir, Syeed, S., Iqra, Z., Ahmad, M. A., Rasool, S., Khursheed, A., Shalla, A. H., Ganaie, A. A., & Rather, M. A. (2023). Antibacterial potential of *Thymus linearis* essential oil collected from Wasturwan mountain: A combination of experimental and theoretical studies involving in silico molecular docking simulation of the major compounds against Novobiocin-resistant mutant of DNA Gyrase-B. *Microbial Pathogenesis*, *183*, 106280-106280.

Garrido-Miranda, K. A., Giraldo, J. P., & Schoebitz, M. (2022). Essential oils and their formulations for the control of Curculionidae pests. *Frontiers in Agronomy, 4.*

Globe Newswire. (2022, October 7). The global essential oils market size is estimated to be valued at USD 8.8 billion in 2022 and is projected to reach USD 15.3 billion by 2027, recording a CAGR of 11.8%. https://www.globenewswire.com/news-release/2022/10/07/2530362/0/en/The-global-essential-oils-market-size-is-estimated-to-be-valued-at-USD-8-8-billion-in-2022-and-is-projected-to-reach-USD-15-3-billion-by-2027-recording-a-CAGR-of-11-8.html

Guzmán, E., & Lucia, A. (2021). Essential oils and their individual components in cosmetic products. *Cosmetics*, *8*(4), 114.

Han, F., Ma, G., Yang, M., Yan, L., Xiong, W., Shu, J., Zhao, Z., & Xu, H. (2017). Chemical composition and antioxidant activities of essential oils from different parts of the oregano. *Journal of Zhejiang University Science B*, *18*(1), 79-84.

Jagannath N., Ramakrishnaiah H., Krishna V, Gowda P. J. (2012). Chemical composition and antimicrobial activity of essential oil of *Heracleum rigens*. *Natural Products Communications*, *7*(7), 943-946.

Jugreet, S., Zengin, G., Zengin, G., & Maggi, F. (2021). Essential oils as natural sources of fragrance compounds for cosmetics and cosmeceuticals. *Molecules*, *26*(3), 666.

Kabdal, T., Himani, Kumar, R., Prakash, O., Nagarkoti, K., Rawat, D. S., Srivastava, R. M., Kumar, S., & Dubey, S. K. (2022). Seasonal variation in the essential oil composition and biological activities of *Thymus linearis* Benth. collected from the Kumaun region of Uttarakhand, India. *Biochemical Systematics and Ecology*, *103*, 104449.

Kharal, D. K., Thapa, U. K., St. George, S., Meilby, H., Rayamajhi, S., & Bhuju, D. R. (2017). Tree-climate relations along an elevational transect in Manang Valley, Central Nepal. *Dendrochronologia*, *41*, 57-64.

Kumar, V., & Mathela, C. S. (2018). Chemical constituents of essential oils of Himalayan *Nepeta ciliaris* Benth. and *Senecio nudicaulis* Buch-Ham. Ex D. Don. *Journal of Essential Oil Research*, *30*(3), 207-213.

Kumar, V., Mathela, C. S., Kumar, M., & Tewari, G. (2019). Antioxidant potential of essential oils from some Himalayan Asteraceae and Lamiaceae species. *Medicine in Drug Discovery*, *1*, 100004.

Liang, J.-Y., Guo, S.-S., Zhang, W.-J., Geng, Z.-F., Deng, Z.-W., Du, S.-S., & Zhang, J. (2017). Fumigant and repellent activities of essential oil extracted from *Artemisia dubia* and its main compounds against two stored product pests. *Natural Product Research*, *32*(10), 1234-1238.

Lohani, H., Haider, S. Z., Chauhan, N. K., & Mohan, M. (2010). Essential oil composition of leaves and berries of *Juniperus communis* and *Juniperus indica* from Uttarakhand Himalaya. *Journal of Medicinal and Aromatic Plant Sciences*, *32*, 199-201.

Mohamed, A. A., & Alotaibi, B. M. (2022). Essential oils of some medicinal plants and their biological activities: A mini review. *Journal of Umm Al-Qura University for Applied Sciences*, *9*(1), 40-49.

Mustafavi, H. S., Abbasi, A., Morshedloo, M. R., Pateiro, M., & Lorenzo, J. M. (2022). Essential oil variability in Iranian populations of *Heracleum persicum* Desf. ex Fischer: A rich source of hexyl butyrate and octyl acetate. *Molecules*, *27*(19), 6296-6296.

Nerio, L. S., Olivero-Verbel, J., & Stashenko, E. E. (2010). Repellent activity of essential oils: A review. *Bioresource Technology*, *101*(1), 372-378.

Observatory of Economic Complexity. (n.d.). Essential oils in Nepal. https://oec.world/en/profile/bilateral-product/essential-oils/reporter/npl

Poudel, K. (2022, July 11). Demand for essential oils in the world is growing. Nepal can benefit from that. The Farsight Nepal. https://farsightnepal.com/news/76#:~:text=Though%20small%2C%20essential%20oils%20exports,most%20exported%20product%20from%20Nepal.

Pradhan, S., Paudel, H. R., Maharjan, R., & Sharma, K. (2023). Essential oils from six aromatic plants of Langtang National Park: Insights on their chemical constituents via GC-MS analysis. *Separations*, *10*(1), 52.

Samarth, R. M., Samarth, M., & Matsumoto, Y. (2017). Medicinally important aromatic plants with radioprotective activity. *Future Science OA*, *3*(4), FSO247.

Satyal, P., Paudel, P., Kafle, A., Pokharel, S. K., Lamichhane, B., Dosoky, N. S., Moriarity, D. M., & Setzer, W. N. (2012). Bioactivities of volatile components from Nepalese *Artemisia* species. *Natural Product Communications*, *7*(12), 1651-1658.

Steiner, J. F., Gurung, T. R., Joshi, S. P., Koch, I., Tuomo Saloranta, Shea, J., Shrestha, A. B., Stigter, E., & Immerzeel, W. W. (2021). Multi year observations of the high mountain water cycle in the Langtang catchment, Central Himalaya. *Hydrological Processes*, *35*(5).

Teixeira, B., Marques, A., Ramos, C., Serrano, C., Matos, O., Neng, N. R., Nogueira, J. M. F., Saraiva, J. A., & Nunes, M. L. (2013). Chemical composition and bioactivity of different oregano (*Origanum vulgare*) extracts and essential oil. *Journal of the Science of Food and Agriculture*, *93*(11), 2707-2714.

Appendix 1 : Qualitative composition (area %) of monoterpenes in the essential oils from the aromatic plant species of the Manang region

S.N.	Compound Name	Composition Area (%)								
		Artemisia dubia	*Elsholtzia fruticosa*	*Tetrataenium lallii*	*Juniperus indica*		*Nepata ciliaris*		*Origanum vulgare*	*Thymus linearis*
					Berry	Leaves	Leaves	Flower		
1	(-) - Globulol		0.8							
2	(-)-*β*-bourbonene			0.4					0.6	
3	(+)-epi-Bicyclosesquiphellandrene	1.3								
4	1-octen-3-ol		1.7						0.9	
5	2-((2R, 4aR,8aS)-4a-Methyl-8-methylenedecane	1.9								
6	2(1H) Naphthalenone,3,5,6,7,8,8a-hexahydro	1.5								
7	2-(4a,8-Dimethyl-2,3,4,5,6,8a-hexahydro-1H)	1.4								
8	3-carene				0.5					
9	4-(1,5-Dimethylhex-4-enyl) cyclohex-2-enone						5.9	8.9		
10	Abietadiene				1.1					
11	Abietal <4-epi->				2.6					
12	Acetic acid, octyl ester			15.7						
13	Acetoxyelemol <8-*α*->					2.8				
14	Androstan-17-one, 3-ethyl-3-hydroxy	2.5								
15	Benzaldehyde		0.3							
16	Bergamotene <*α*-, cis>						5.2	5.8		
17	Bergamotene <*β*-, trans>						1.0	1.4		
18	Bornyl acetate			8.6						
19	Butanoate <hexyl-, 3-methyl->			6.6						
20	Butanoic acid, 2-methyl-, 2-methylbutyl ester			2.5						
21	Butanoic acid, 2-methyl-, hexyl ester			4.8						
22	Butanoic acid, 3-methyl-, 3-methylbutyl ester			1.6						
23	Butyrate <butyl-, 2-methyl->			3.2						
24	Butyrate <hexyl>			1.4						
25	Butyrate <isobutyl-, 2-methyl->			1.5						
26	Cadin-4-en-10-ol		0.6							
27	Carvacrol									6.2
28	Carvacryl methyl ether								4.5	
29	Caryophyllene oxide	1.9					29.0	34.4		
30	Chamazulene	6.7								
31	Cis-chrysanthenol	3.8								
32	Citronellate <methyl->					0.5				
33	Copaene	1.9								
34	Curcumen-15-al <ar->							1.1		
35	Decanal <n->			0.9						
36	D-Limonene	2.7	2.9	15.4	23.9	28.5				

S.N.	Compound Name	Composition Area (%)								
		Artemisia dubia	*Elsholtzia fruticosa*	*Tetrataenium lallii*	*Juniperus indica*		*Nepata ciliaris*		*Origanum vulgare*	*Thymus linearis*
					Berry	Leaves	Leaves	Flower		
37	E-Caryophyllene	3.5	2.6		0.4		6.0	3.5		0.5
38	E-Nerolidol	3.3	4.3							
39	Ethanone, 1-(1-cyclohexen-1-yl)-						5.3	2.8		
40	Eucalyptol		1.4	2.2		0.6				5.6
41	Eugenol									0.2
42	E-*β*-ocimene		1.5						0.9	
43	Germacrene B				0.1					
44	Germacrene B				1.2					
45	Germacrene D	1.2			0.2				0.5	0.1
46	Hexacosane									
47	Hexanoic acid, hexyl ester			1.2						
48	Hexanoic acid, octyl ester			0.4						
49	Humulene epoxide II							2.4		
50	Isobutyl isovalerate			1.6						
51	Isobutyrate <hexyl->			6.1						
52	Isobutyrate <isobutyl>			0.4						
53	Isopimara-9(11),15-diene					0.5				
54	Isovalerate <butyl>			3.1						
55	Linalool oxide <cis>								0.3	
56	Linalyl anthranilate	2.9		1.1		0.5		0.5		
57	Naphthalene, decahydro-4a-methyl-1-methyl	6.9								
58	Napth-1-ol < 1,2,3,4,4a,7,8,8a-octahydro-,4				0.5					
59	Norbornane < 2,2-dimethyl-,5-methylene->									0.1
60	n-pentacosane									
61	Octan-3-ol								0.6	0.3
62	Octan-3-one								2.9	2.1
63	p-cymen-8-ol								0.8	
64	p-cymene	4.2	9.6	5.3	0.4				34.1	12.4
65	Pregeijerene B					1.6				
66	Propionic acid, 2-methyl-, 3-methylbutyl ester			0.9						
67	Sabinene		0.6		20.2	15.8				0.3
68	Sabinene hydrate <trans>		4.6							2.0
69	Santolina triene									
70	Sesquisabinene						10.8	5.8		0.2
71	Terpinen-4-ol	6.0	18.1		3.1	7.3		0.4	0.8	1.0
72	Terpinolene	1.2								
73	Thymol			0.6					10.4	41.3
74	Thymol acetate									6.7

S.N.	Compound Name	Composition Area (%)								
		Artemisia dubia	*Elsholtzia fruticosa*	*Tetrataenium lallii*	*Juniperus indica*		*Nepata ciliaris*		*Origanum vulgare*	*Thymus linearis*
					Berry	**Leaves**	**Leaves**	**Flower**		
75	Thymol methyl ether								12.1	0.9
76	Totarol <trans>					0.4				
77	Valeranone								1.8	
78	Z-α-bisabolene									0.1
79	Z-β-ocimene		1.0						0.5	
80	α - epi Muurolol	2.1								
81	α - phellandrene	2.9	0.7							0.2
82	α - terpniene geranyl	4.0								
83	α -pinene		1.1	2.4	5.6	2.6			0.7	1.5
84	α -terpinene	3.3	8.0		1.0	1.8			1.1	1.4
85	α -terpineol		3.8	0.5						1.0
86	α -terpinolene		3.8		1.5	1.5				0.2
87	α-copaene	2.4					0.9	0.5		
88	α-curcumene						0.9	1.3		
89	α-elemol	2.1			9.5	10.9				
90	α-humulene						0.6			
91	α-muurolene	1.2								
92	α-thujene		2.4		2.5	1.5			1.1	2.5
93	β - oplopenone					0.7				
94	β-bisabolene						3.0	3.5	4.5	5.8
95	β-elemene				0.2					
96	β-eudesmol	3.8			2.0					
97	β-myrcene		0.2		2.4	2.0			1.2	0.5
98	β-pinene									1.1
99	β-selinene									
100	γ- epi Eudesmol					0.5				
101	γ-eudesmol		1.3		2.5	2.2				
103	γ-terpinene	9.3	16.5	6.4	1.9	2.9			6.9	5.9
104	δ-3-carene									0.2
105	δ-cadinene	3.2	1.3		0.7	0.8				
106	τ-cadinol		0.8							
	Total	88.9	89.8	94.7	86.3	88.0	68.6	72.2	87.1	100.0

Journal of Plant Resources (2025) **23**(1), 45-56
https://doi.org/10.3126/bdpr.v23i1.83350

Antimicrobial Activity of Essential Oil from *Tagetes minuta* L. and its Extracts along with its GC-MS Profiling

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Abstract

Nepal, being a land of diverse topography with the different weather conditions, is considered as storehouse of medicinal and aromatic plants (MAPs) with enriches medicinal values. *Tagetes minuta* L. is highly demanded aromatic plant for its essential oil, hugely used in flavor and perfumery industries, having potential bioactive and therapeutic properties. The essential oil was extracted from aerial parts of *Tagetes minuta L.* via hydro-distillation and its chemical composition was analyzed using GC-MS technique. Furthermore, the antimicrobial activity of oil and its hexane, ethylacetate, and methanolic extracts was assessed against different pathogenic bacterial strains using agar well diffusion method. The essential oil yield was 0.3% (v/w) and GC-MS data revealed the highest percentage of monoterpenes: alpha pinene (33.12 %), transocimenone (23.32 %), cistagetone (12.18%) and dihydrotagetone (9.65%) along with other hydrocarbons. Similarly, the antimicrobial activity showed ethylacetate and methanolic extracts to be quite active against pathogenic bacterial strains *Staphylococcus aureus* (ATCC 6538P) with Zones of Inhibition (ZOI) 14.30 mm and 10.40 mm and *Staphylococcus epidermidis* (ATCC 12228) with ZOI value of 17.50 mm and 16.30 mm respectively. On the other hand, it also showed moderate activity against pathogenic bacterial strains *Bacillus subtilis* (ATCC 8739) and *Proteus vulgaris* (ATCC 6380*)*. Although additional necessary investigations are needed, these results support the use of traditional medicinal plants in treating various diseases. Also, these results suggest that the studied essential oil may have potential for isolating the bioactive compounds, which could contribute to the discovery of noble drugs in future.

Keywords: Medicinal and aromatic plants, Monoterpenes , α-pinene, *Tagetes minuta* L. essential oil

Introduction

Nepal is ranked as 9th among the Asian countries for its floral wealth with an estimated 9,000 species of flowering plants (Kunwar et al., 2023). Among which, it is supposed that 1600-1900 species are medicinal and aromatic plants commonly used for the medicinal purpose (Ghimire et al., 2008).

Essential oils (EOs), also referred as ethereal or volatile oils, have been used for extensive applications in pharmaceutical, medical, and perfume industries. They are aromatic oily liquids obtained from different parts of plant, i.e., leaves, seeds, fruits, buds, flowers, herbs, barks, and roots (Silori et al., 2019). Similarly, they are natural extracts of aromatic plants used in many fields like agriculture, aromatherapy and nutrition (Bolouri et al., 2022). EOs are a complex mixture of mainly terpenes particularly monoterpenes and sesquiterpenes and their oxygenated derivatives such as alcohols, aldehydes, esters, ethers, ketones, phenols, and oxides (Badawy & Abdelgaleil, 2014). The pharmacological studies suggest that these EOs can be used as anti-rheumatic, antiseptic, antispasmodic, anticancer, anti-inflammatory, antitoxic, aphrodisiac, and astringent agents (Bhardwaj et al., 2020).

Tagetes minuta L. is annual, strongly aromatic herb of sunflower family (Asteraceae), one of the most abundant plants taxonomical grouping, comprising about 1,000 genera and over 23,000 species (Sadia et al., 2013).The genus *Tagetes* refers to the Latin name for marigold '*Tages*', an Etruscan god associated with agriculture and the species *T. minuta* is from the Latin word '*minutes*' meaning small referring to its small sized capitula (Bandana et al., 2018). The stem is 1-2 meter tall and branched with 5-15 cm long opposite leaves divided into one terminal and several (3-7) lateral leaflets. Flowers are creamy yellow in color and blooms from September to December in late summer (Hulina, 2008). This plant has been initially originated from South America and found in temperate grasslands and elevated regions.

In the context of Nepal, *T. minuta* has been listed as an medicinal plants prioritized for economic development (Department of Plant Resources [DPR], 2024). This plant thrives particularly in western Himalayan region and has gained popularity among farmers for its essential oil production and aroma industries (Walia & Kumar, 2020). This plant exhibits moderate tolerance to various soil conditions, making it suitable for diverse agricultural environments in Nepal. Similarly, its resilience to adverse conditions, such as high salinity and pH levels, enhances its potential as a sustainable crop in challenging terrains (Sahay & Patra, 2013). Walia et al. (2020) performed a recent study on variations in essential oil composition of *T. minuta* L. provided the meaningful information for diversified phytotherapy and for improved utilization in food, flavor and fragrance industries. Extensive studies on the phytochemistry and pharmacology of *T. minuta* have identified the presence of essential oils, thiophenes, flavonoids, terpenes, saponins, carotenoids, and other chemical compounds in this species (Verma et al., 2024). Meshkatalsadat et al. (2010) conducted a research on the chemical characterization of volatile components of *T. minuta* cultivated in south west of Iran by nano scale injection technique confirmed the presence of more than 27 compounds that are mainly limonene, piperitenone, α-terpinolene, piperitone, (E)-tagetone, (Z)-ocimenone and so on. Similarly, another research performed by Singh et al. (2016) identified the major chemical constituents to be Z-β-ocimene, limonene, dihydrotagetone, tagetones (E&Z) and ocimenones (E & Z).

In recent years, a wide range of plant essential oils and their constituents have been investigated for their antibacterial properties against an array of plant pathogenic bacteria (Gakuubi et al., 2016) and they have also several medicinal benefits, which include remedy for colds, respiratory inflammations and stomach problems (Karimian et al., 2014a). Another research emphasized naturalized exotic weed, *Tagetes minuta* as a potentially invasive species in Nepal (Lamichhane et al., 2025). Some previous studies performed in Nepal on this plant have highlighted its antimicrobial properties as well as its ethno-medicinal values (Khakurel et al., 2014). Although a few articles on the antimicrobial studies of essential oils obtained from this plant have previously been reported internationally (Senatore et al., 2004) and nationally (Joshi et al., 2014), the antimicrobial activities of various Soxhlet extracts have not yet been thoroughly explored, particularly in the context of Nepal. The major aim of the present study was to evaluate the antimicrobial activities of hexane, ethyl acetate and methanolic extracts of *T. minuta* as well as its essential oil, against different pathogenic bacterial and fungi strains.

Materials and Methods

Sample collection

The fresh plants of *T. minuta* (collection no. M003) were collected from Marpha of Mustang district in the month of October, 2023. The locality where the plant material was collected was nearly at the altitude of 2700 meter above the sea level. The plant material was identified by National Herbarium and Plant Laboratories (KATH), Godawari, Lalitpur with the voucher code no. KATH170291.

Essential oil extraction

Essential oil from the shade-dried plant sample was extracted by hydro-distillation method. Small pieces of aerial parts of the plant materials were tightly packed in round bottom flask and fitted to a Clevenger apparatus and condenser. The oil was extracted for about 4 hours and was collected in glass vial. The moisture present in the essential oil was later removed by using anhydrous sodium sulphate and the obtained oil was stored under refrigerated conditions (Walia et al., 2020).

The essential oil content (v/w) % was calculated using the formula:

$$\text{Essential oil content (\%)} = \frac{E_1}{E_2} \times 100$$

Where, E_1= Essential oil volume (mL),
E_2= Fresh sample weight (gm)

Identification of compounds of essential oil by GC-MS

The essential oil obtained from *T. minuta* was dissolved in solvent forming 1:10 ratio i.e. the sample solution was prepared in a sample vial by dissolving 100 µL of essential oil in 1000 µL hexane as solvent, sonicating for 5-10 minutes and then used for further process.

Compounds of *T. minuta* essential oil were analyzed by Shimadzu GC-MS-QP 2010 Plus available at the Instrument Section of the Department of Plant Resources. The capillary column used for the analysis was SH-RTX-5MS (60 m _ 0.32 mm _ 0.25 m) with a crossbond of 5% diphenyl / 95% dimethyl polysiloxane as the stationary phase. The GC analysis was performed under the following conditions: column oven temperature - 50°C, injection temperature - 250°C, ion source temperature - 250°C, interface temperature - 200°C, split injection mode with a split ratio of 80, electron impact (EI) mode - 70 eV, ion source temperature - 250°C, Helium with a pressure of 53.8 kPa, total gas flow - 112.3 mL/min and column flow - 1.35 mL/min. The GC-MS system starts with an initial oven temperature of 50°C for 1 minute, and then increases to 230°C at a rate of 3°C for 9 minutes. Mass spectral detection was carried out in electron ionization mode by scanning at 40 to 350 m/z. The total time required for analyzing a single sample was 70 min (Pradhan et al., 2023).

The chemical components of the essential oils were identified by comparing their mass spectral fragmentation patterns with those in the National Institute of Standard Technology Library (NIST) 2017 and Flavor and Fragrance Natural and Synthetic Compounds FFNSC 4.0 library search facility linked with the data analysis software of the GC-MS (Adams, 2005). The percentage of each component (Area %) is reported as raw percentages based on the total ion chromatogram (TIC) without standardization.

Soxhlet extraction method

The crude sample was placed in a thimble-shaped filter paper which is then kept in a glass cylinder provided with a siphon tube and an inlet tube. A water condenser was attached to the cylinder at the top and the entire assembly was fitted into the neck of a round bottom flask containing the solvent. The flask was heated in water bath. The solvent vapors reached the cylinder through the inlet tube and condensed on passing upward into the condenser. The condensed solvent came in contact with the crude organic substance and dissolved it as soon as the solution reached the top end of the siphon tube. In this way, a continuous supply of solvent vapors was maintained in the cylinder, and the dissolved organic compound flowed back into the flask. Finally, the heating was stopped and the solution in the flask was distilled to recover the solvent, while the organic compound left behind was collected as the form of sample extract.

Here, the Hexane (non-polar), ethylacetate (moderately polar) and methanol (polar) solvents were consecutively used as the solvents (Abubakar & Haque, 2020).

Antimicrobial activity test by agar well diffusion method

The antimicrobial activity of essential oils of *T. minuta* was evaluated by agar well diffusion method (AWD) as described by Clinical and Laboratory Standard Institute (CLSI) guidelines (Cockerill & CLSI, 2012). The pathogenicity of the bacterial isolates were not checked and the used bacterial strains (Microbiologics, Cooper Avenue North, Saint cloud, Minnesota, USA) were of: *Bacillus subtilis* (ATCC 6059), *Enterococcus faecalis* (ATCC 29212), *Escherichia coli* (ATCC 8739), *Klebsiella quasipneumonia* (ATCC 700603), *Proteus vulgaris* (ATCC 6380), *Pseudomonas aeruginosa* (ATCC 9027), *Salmonella entericatyphi* (clinical sample), *Shigella dysenteriae* (clinical sample), *Staphylococcus aureus* (ATCC 6538P) and *Staphylococcus epidermidis* (ATCC 12228) and a fungi *Candida albicans* (ATCC 10231).

The bacterial and fungal strains were cultured in nutrient agar and incubated at 35 °C in incubator (Accumax Equipments, India) for growth. The inoculum suspension was prepared in normal saline solution and was spread uniformly with a sterile cotton swab on Mueller Hinton Agar (HiMedia, India) plates. A 6 mm diameter hole was aseptically punched on the agar surface with a sterile cork borer, and 50 µL of the essential oil/sample extract were introduced into each well. Ciprofloxacin (CIP 5 mcg/disc) were used as positive control for tested bacterial strains- *B. subtilis, E. faecalis, E. coli, K. quasipneumoniae, P. vulgaris, P. aeruginosa, S. typhi and S. dysenteriae,* Amoxycillin (AMX 10 mcg/disc) for *S. aureus and S. epidermidis* and Clotrimazole (CL 10 mcg/disc) for fungi *C. albicans* respectively. Similarly, dimethyl sulfoxide (DMSO) was used as negative control. The plates were kept for 2-3 hours to allow diffusion of essential oils into the agar medium and incubated at 35°C for 24 hours. In this way, antimicrobial assay was performed and zone of inhibition were measured by using vernier caliper. The sensitivity was categorized as: not sensitive (diameter ≤8 mm); sensitive (diameter 9-14 mm); very sensitive (≥15 mm) (Ponce et al., 2003).

Results and Discussion

Essential oil content and composition

A yellow colored essential oil was obtained from the fresh biomass of *Tagetes minuta* L. yielding 0.3% (v/w) based on their fresh weight. Previous studies of essential oil from four *Tagetes* species collected in Hungary revealed the oil percentage ranged between (0.5-1.18)% (Hethelyi, et al., 1986). Bahadirli (2020) reported that 1.8% of essential oil was isolated from *Tagetes minuta* from Turkey. Similarly, Walia et al. (2020) reported that the essential oil content of fresh *T. minuta* plant varied from 0.37 to 0.79% across different altitudinal locations.

The chemical composition of the essential oil was analyzed using Gas Chromatography-Mass Spectroscopy (GC-MS), which enabled the identification of several major constituents contributing predominantly to the total oil content. GC-MS analysis of the essential oil led to the identification of over 20 distinct volatile compounds in the essential oil. Among these, α-pinene exhibited the highest relative abundance, accounting for 33.12% of the total peak area, followed by trans-ocimenone (23.32%). Other prominent constituents included cis-tagetone (12.18%), dihydrotagetone (9.65%), and cis-ocimenone (7.91%). The percentage composition of monoterpene hydrocarbon, dihydrotagetone, ocimenone is found to be comparable with previous study reported by (Babaei et al., 2021). Their research revealed that water limitation had a considerable effect on the essential oil composition of *T. minuta*. Results showed that out of 30 compounds, the essential oil mainly consisted of oxygenated monoterpenes (65.3-75.3%), which represented with dihydrotagetone (35.8-40.9%) as the major component, followed by *E*-(Z)-tagetone (15.4-17.6%). Monoterpene hydrocarbons (20.6-27.2%) represented by β-ocimene (11.6-18.5%) were also identified along with other components (E)-ocimenone, (Z)-ocimenone, limonene and so on. GC-MS analysis of essential oil of *T.minuta* L. revealed a greatly diversified proportions of compounds rich in many secondary compounds, including monocyclic and bicyclic monoterpenes (47.90%), sesquiterpenes (30.20%), hemiterpenes (15.13%) and diterpenes (1.68%), confirmed from the previous findings by Wanzala & Ogoma (2013). Their extensive research highlighted the higher proportion of monoterpene constituents of *Tagetes* oil compared favourably with the results obtained from this report. The results from this study and those from other literature further confirmsthat the main principal constituents of the essential oil of *T. minuta* are ocimene, dihydrotagetone, tagetones and ocimenones. These compounds form the basis of using this oil in the pharmaceutical, agricultural, food and perfumery industries, contributing its high demand (Singh et al., 2003).

α-Pinene is a bicyclic monoterpene widely found naturally as an insect-repellent agent in plant defense (Huang et al., 2013). When α- and β-pinenes are the major constituents of an essential oil, they warrant the anti-inflammatory and analgesic activity (Mercier et al., 2009). Similarly, ocimenone, an unsaturated

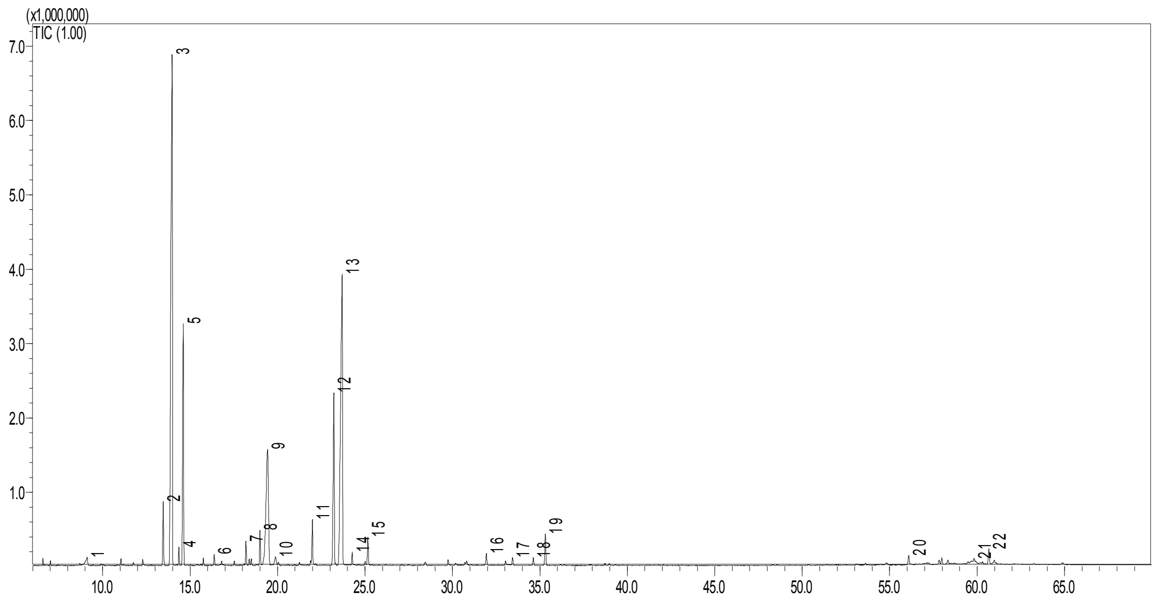


Figure 1: Chromatogram of *Tagetes minuta* L. essential oil representing Retention time (min) in X-axis vs Intensity (TIC * 1,000,000) in Y-axis

acylicmonoterpene hydrocarbon and tagetone and dihydrotagetone, oxygenated monoterpene, are also present abundantly in *T. minuta* essential oil similar as reported from Kyarimpa et al. (2015) as well as Rathore et al. (2018). Extensive studies on the phytochemistry has revealed the presence of various phytoconstituents such as thiophenes, flavonoids, terpenes, saponins, carotenoids, and other chemical compounds in its essential oil. These phytoconstituents contribute to various pharmacological activities, including antimicrobial (Al-Robai et al., 2023), anticancer (Oyenihi et al., 2021), antidiabetic (Lim et al., 2023), antioxidant and anti-inflammatory properties (Karimian et al., 2014b) etc.

The GC-MS chromatogram and the corresponding total ion chromatogram (TIC) profile are presented in (Figure 1), illustrating the separation and relative abundance of the detected compounds. The components, identified based on their mass spectral data, and retention time are listed in (Table 1).

Table 1: Chemical composition of *Tagetes minuta* L. essential oil

Peak	Retention Time (min)	Area %	Name
1.	9.116	0.8	3,3,5-Trimethylcyclohexene
2.	13.462	2.13	Limonene
3.	13.977	33.12	α-Pinene
4.	14.359	0.58	(E)-β-Ocimene
5.	14.61	9.65	Dihydrotagetone
6.	16.38	0.38	Carvenone
7.	18.193	0.84	neo-allo-Ocimene
8.	18.994	1.23	trans-Tagetone
9.	19.428	12.18	cis-Tagetone
10.	19.884	0.64	2-allylfuran-3-carboxylate
11.	21.99	1.65	4-methyleneisophorone
12.	23.221	7.91	cis-Ocimenone
13.	23.695	23.32	trans-Ocimenone
14.	24.268	0.48	Carvacrol
15.	25.162	1.11	Livacone
16.	31.938	0.56	(E)-Caryophyllene
17.	33.435	0.34	α-Humulene
18.	34.623	0.31	γ-Cadinene
19.	35.31	1.3	Bicyclogermacrene
20.	56.086	0.47	trans-α-Atlantone
21.	59.822	0.36	Adamantan-2-one
22.	60.668	0.66	Artemisia ketone

Antimicrobial activity

The results on antimicrobial activity test of essential oil of *T. minuta* against different pathogenic bacterial strains are tabulated in (Table 2). The essential oil of *T. minuta* demonstrated promising antimicrobial activity against the gram-negative bacterial strain *Proteus vulgaris*, and the gram positive bacterial strains *Staphylococcus aureus* and *Staphylococcus epidermidis*, showing significant zone of inhibition of 10.50 mm, 12.30 mm and

11.20 mm respectively. This can be interpreted as it was moderately active as compared to the positive controls used i.e. Ciprofloxacin with zone of inhibition 32.06 mm and Amoxycillin with ZOI 35.10 mm and 17.74 mm. Senatore et.al. (2004) evaluated the antimicrobial activity of essential oil of *T. minuta* against different gram-positive and gram-negative bacterial strains using broth dilution method. Their findings suggested that UK (United Kingdom) based sample oil was more active than the South African samples against all the bacteria tested and more active than the Egyptian oil against all bacterial strains except the gram-negative *Proteus mirabilis*, against which it showed the same activity. Gakuubi et al. (2006) revealed that the essential oils of *T. minuta* showed promising antibacterial activities against the test pathogens with *Pseudomonas savastanoi* pv. *Phaseolicola* among others (*Xanthomonas axonopodis* pv. *Phaseol* and *Xanthomonas axonopodis* pv. *manihotis*) being the most susceptible with mean inhibition zone diameters of 41.83 mm and 44.83 mm after 24 and 48 hours, respectively. The minimum inhibitory concentrations and minimum bactericidal concentrations of the EOs on the test bacteria were in the ranges of 24-48 mg/mL and 95-190 mg/mL, respectively. Similarly, Walia et al. (2020) investigated the antimicrobial activity against two gram-positive bacteria viz. *Micrococcus luteus*, and *Staphylococcus aureus*, and two gram-negative bacteria viz. *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* using agar well-diffusion and micro-dilution methods. The agar well diffusion method demonstrated stronger activity of *T. minuta* EOs in gram positive bacteria as compared to gram negative bacteria. The highest activity was observed against *S. aureus* with zone of inhibition above 9 mm. The most potential EOs (three locations of Himalchal Pradesh and one location of Manipur) demonstrated an MIC of 25-30% (v/v). According to Trombetta et al. (2005), gram-positive bacterial strains are usually more susceptible to the antibacterial activity of lipophilic essential oils because of the lipophilic nature of the gram-positive cell membrane, as compared to the hydrophilic character of the cell membrane component of gram-negative bacteria.

Different Soxhlet extracts of *T. minuta* exhibited some antimicrobial activity against various tested bacterial strains as tabulated in (Table 3). Among the tested three extracts, hexane extract did not show any activity against all the tested microbes, but ethyl acetate and methanolic extracts showed significant activity against different bacterial strains. Hexane, being a non-polar solvent does not efficiently extract polar or semi-polar phytochemicals such as

Table 2: Antimicrobial activity test of essential oil of *T. minuta* L

S.N.	Microorganism	Reference culture	Test extract (*T. minuta* L. essential oil)	ZOI of Solvent (mm)	Positive control		
			Zone of Inhibition (ZOI) (mm)		Name	Conc. (mcg)	ZOI (mm)
	Bacteria						
1.	*Bacillus subtilis*	ATCC 6051	0.00	0.00	Ciprofloxacin	5	31.16
2.	*Enterococcus faecalis*	ATCC 29212	0.00	0.00	Ciprofloxacin	5	21.40
3.	*Escherichia coli*	ATCC 8739	0.00	0.00	Ciprofloxacin	5	32.38
4.	*Klebsiella quasipneumoniae*	ATCC 700603	0.00	0.00	Ciprofloxacin	5	23.60
5.	*Proteus vulgaris*	ATCC 6380	10.50	0.00	Ciprofloxacin	5	32.06
6.	*Pseudomonas aeruginosa*	ATCC 9027	0.00	0.00	Ciprofloxacin	5	30.28
7.	*Salmonella enterica* subsp. *enterica* pv Typhi	Clinical Sample	0.00	0.00	Ciprofloxacin	5	34.60
8.	*Shigella dysenteriae*	Clinical Sample	0.00	0.00	Ciprofloxacin	5	33.72
9.	*Staphylococcus aureus*	ATCC 6538P	12.30	0.00	Amoxycillin	10	35.10
10.	*Staphylococcus epidermidis*	ATCC 12228	11.20	0.00	Amoxycillin	10	17.74
	Fungi						
1.	*Candida albicans*	ATCC 10231	0.00	0.00	Clotrimazole	10	16.20

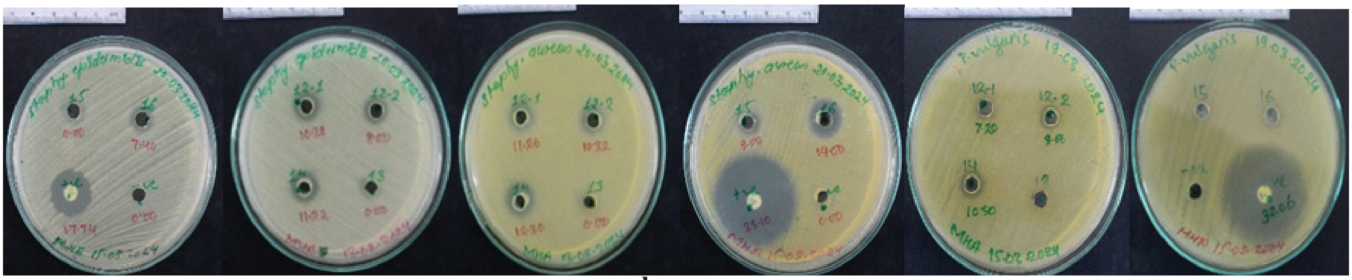
flavonoids, phenolic, alkaloids which are often most associated with the antimicrobial activity (Verma et al., 2024). A key study isolated 19 acylated flavonol glycosides all from butanol and ethylacetate extracts that showed significant antibacterial activity. In contrast, the hexane fraction did not contain these active flavonols (Shahzadi & Shah, 2015). The ethylacetate extract revealed noticeable activity against *Bacillus subtilis*, *Staphylococcus aureus*, *Staphylococcus epidermidis* with ZOI of 10.10 mm, 14.30 mm, and 17.50 mm respectively. Similarly, the methanolic extract showed a promising activity against *Proteus vulgaris*, *Staphylococcus aureus*, *Staphylococcus epidermidis* with ZOI of 9.80 mm, 10.40 mm and 16.30 mm respectively. Pillai et al. (2020) investigated the antimicrobial activity of *T. minuta* stem bark extracts obtained via maceration using four different solvents: hexane, ethylacetate, methanol and chloroform. Using the hole-plate diffusion method, they tested the extracts against six bacterial isolates viz. *Staphylococcus aureus*, *Listeria monocytogenes*, *Escherichia coli* (wild), *Escherichia coli* (O157:H7), *Pseudomonas aeruginosa* and *Serratia marcescens* and two fungal

Table 3: Antimicrobial activity test of Soxhlet extracts of *T. minuta* L. aerial part

S.N.	Microorganism	Soxhlet extracts of *T. minuta* L. whole shoot plant	Zone of Inhibition (ZOI) (mm)	ZOI of Solvent (mm)	Positive control		
					Name	Conc. (mcg)	ZOI (mm)
	Bacteria						
1.	*Bacillus subtilis*	Hexane	0.00	0.00	Ciprofloxacin	5	31.16
		Ethyl Acetate	10.10				
		Methanol	0.00				
2.	*Enterococcus faecalis*	Hexane	0.00	0.00	Ciprofloxacin	5	21.40
		Ethyl Acetate					
		Methanol					
3.	*Escherichia coli*	Hexane	0.00	0.00	Ciprofloxacin	5	32.38
		Ethyl Acetate					
		Methanol					
4.	*Klebsiella quasipneumoniae*	Hexane	0.00	0.00	Ciprofloxacin	5	23.60
		Ethyl Acetate					
		Methanol					
5.	*Proteus vulgaris*	Hexane	0.00	0.00	Ciprofloxacin	5	32.06
		Ethyl Acetate	0.00				
		Methanol	9.50				
6.	*Pseudomonas aeruginosa*	Hexane	0.00	0.00	Ciprofloxacin	5	30.28
		Ethyl Acetate					
		Methanol					
7.	*Salmonella enterica* subsp. *enterica* pv Typhi	Hexane	0.00	0.00	Ciprofloxacin	5	34.60
		Ethyl Acetate					
		Methanol					
8.	*Shigella dysenteriae*	Hexane	0.00	0.00	Ciprofloxacin	5	33.72
		Ethyl Acetate					
		Methanol					
9.	*Staphylococcus aureus*	Hexane	0.00	0.00	Amoxycillin	10	35.10
		Ethyl Acetate	14.30				
		Methanol	10.40				
10.	*Staphylococcus epidermidis*	Hexane	0.00	0.00	Amoxycillin	10	17.74
		Ethyl Acetate	17.50				
		Methanol	16.30				
	Fungi						
1.	*Candida albicans*	Hexane	0.00	0.00	Clotrimazole	10	16.20
		Ethyl Acetate					
		Methanol					

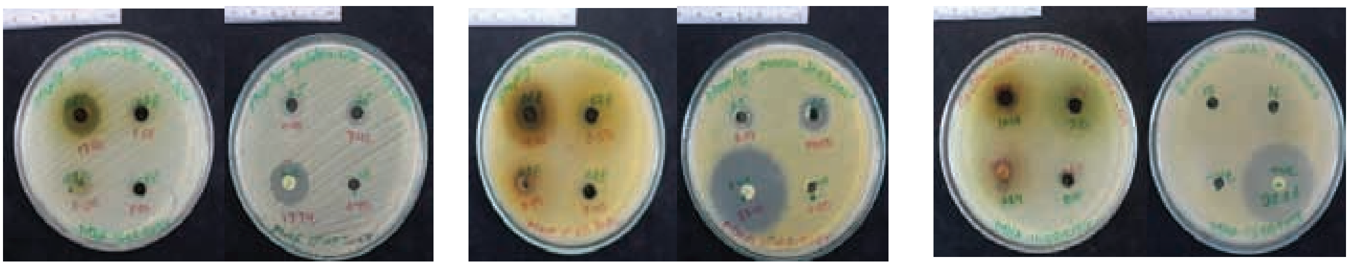
isolates viz. *Candida albicans* and *Penicillium digitatum*. The inhibition zones were found to be in the ranges of 10.0±1.6 to 15.5±1.9 mm against bacterial isolates and 11.3±2.1 to 13.4±1.2 mm against *P. digitatum* but no activity was observed against *C. albicans* aligning the result of the present study. Therefore, *T. minuta*, being a very valuable plant, may also be useful for the treatment of diseases caused by different human pathogens. Such as: boils, blisters and redness of skin due to skin and soft skin infections by *S. Aureus* (Tong et al., 2015); sinus infection, endocarditis, intravascular catheter infections, cardiac devices, prosthetic joints, and CNS shunt infection by *S. epidermidis* (Lee & Anjum, 2024); urinary tract infection, wound infection, skin infection and respiratory tract infection caused by *P. vulgaris* (Kim et al., 2003) and infection of the blood, heart, lung, bone, eye, and brain infections by *B. subtilis* (Tokano et al., 2023), among others.

A picture showing positive zone of inhibition of *T. minuta* EO and its extracts (ethylacetate and methanolic) against different pathogens along with its positive and negative controls are depicted in Figure 2, Figure 3 and Figure 4 for further broader analysis.



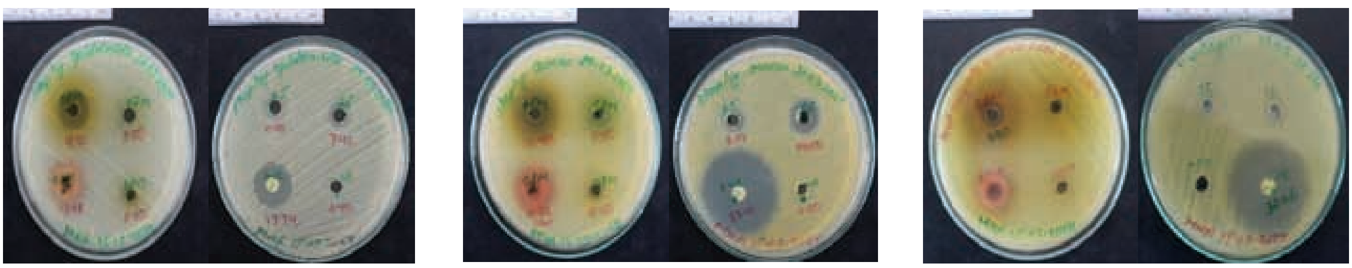
a. **b.** **c.**

Figure 2: Inhibition zones of *T. minuta* L. essential oil i.e. sample no. 14 against pathogenic strains, (**a.** *Staphylococus epidermidis*, **b.** *Staphylococcus aureus*, **c.** *Proteus vulgaris*) with their adjoining positive and negative controls



a. **b.** **c.**

Figure 3: Inhibition zones of *T. minuta* L. ethylacetate extract i.e. 06E against pathogenic strains (**a.** *Staphylococus epidermidis*, **b**. *Staphylococcus aureus*, **c.** *Bacillus subtilis*) with their adjoining positive and negative controls



a. **b.** **c.**

Figure 4: Inhibition zones of *T. minuta* L. methanolic extract i.e. 06M against pathogenic strains (**a.** *Staphylococus epidermidis*, **b.** *Staphylococcus aureus*, **c.** *Proteus vulgaris*) with their adjoining positive and negative controls

Conclusion

The essential oil extracted from *T. minuta* L. by the hydro-distillation method and analyzed by GC-MS demonstrated a rich chemical profile, with various other bioactive compounds. These constituents highlight the potential application plant in the pharmaceutical, agricultural, food, and perfumery industries. In antibacterial screening, the oil exhibited satisfactory inhibition zones with different bacterial strains most efficiently with gram-positive bacteria, suggesting it as a promising agent for further medicinal development. In addition to, the plant extracts show potential for antibacterial drug discovery and support ethno-pharmacological use and commercialization. But, in order to enlighten more precisely, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) can be suggested furthermore. This study confirms *T. minuta* as a naturally available, cheap, safe, and effective alternative to chemical bactericides. Finally, in the context of Nepal, *Tagetes minuta*, a potential medicinal herb, should be promoted for commercial cultivation, and stakeholders in the Nepalese business community should explore its market potential with continuous efforts and initiatives.

Author contributions

S. Adhikari did sample collection, Soxhlet extraction, antimicrobial analysis and manuscript preparation and S. Aryal also assisted during sample collection and also essential oil extraction, GC-MS analysis and Data recording.

Acknowledgements

The authors would like to thank Dr. Sanjeev Kumar Rai, Director General and Mr. Saroj Kumar Chaudhary, Deputy Director General, of the Department of Plant Resources for their continuous encouragement and guidance. Mr. Chandra Mani Aryal and Ms. Shanta Sapkota are thankful for their co-operation during sample collection, field support and special gratitude. Also, authors are thankful to Mr. Pramesh Bahadur Lakhey for cooperation during antimicrobial activity test analysis. The authors would also like to acknowledge the Department of Plant Resources for providing the opportunity to conduct the research work and laboratory assistance.

References

Abubakar, A. R., & Haque, M. (2020). Preparation of medicinal plants: Basic extraction and Fractionation procedures for experimental purposes. *Journal of Pharmacy & Bio-allied Sciences*, *12*(1), 1-10. https://doi.org/10.4103/jpbs.JPBS_175_19

Adams, D. R. P. (2008). Identification of essential oil components by Gas chromatography. *Allured Publishing Corporation*, *4*, 804.

Al-Robai, S. A., Ahmed, A. A., Ahmed, A. A. E., Zabin, S. A., Mohamed, H. A., & Alghamdi, A. A. A. (2023). Phenols, antioxidant and anticancer properties of *Tagetes minuta*, *Euphorbia granulata* and*Galinsoga parviflora*: *In vitro* and *in silico* evaluation. *Journal of Umm Al-Qura University for Applied Sciences*, *9*(1), 15-28. https://doi.org/10.1007/s43994-022-00017-2

Babaei, K., Moghaddam, M., Farhadi, N., & Ghasemi Pirbalouti, A. (2021). Morphological, physiological and phytochemical responses of Mexican marigold (*Tagetes minuta* L.) to drought stress. *Scientia Horticulturae*, *284*, 110116. https://doi.org/10.1016/j.scienta.2021.110116

Badawy, M. E. I., & Abdelgaleil, S. A. M. (2014). Composition and antimicrobial activity of essential oils isolated from Egyptian plants against plant pathogenic bacteria and fungi. *Industrial Crops and Products*, *52*, 776-782. https://doi.org/10.1016/j.indcrop.2013.12.003

Bahadirli, N. P. (2020). Essential oil content and compositions of naturalized *Tagetes minuta* L. (wild marigold). *Natural Volatiles and Essential Oils*, *7*(2), 17-21. https://doi.org/10.37929/nveo.704056

Bandana, K., Raina, R., Kumari, M., & Rani, J. (2018). *Tagetes minuta:* An overview. *International Journal of Chemical Studies*, 3711-3717.

Bhardwaj, K., Islam, M., Jayasena, V., Sharma, B., Sharma, S., Sharma, P., Kuca, K., & Bhardwaj, P. (2020). Review on essential oils, chemical composition, extraction, and utilization of some conifers in Northwestern Himalayas. *Phytotherapy Research*, *34*. https://doi.org/10.1002/ptr.6736

Bolouri, P., Salami, R., Kouhi, S., Kordi, M., Asgari Lajayer, B., Hadian, J., & Astatkie, T. (2022). Applications of essential oils and plant extracts in different industries. *Molecules*, *27*(24). https://doi.org/10.3390/molecules27248999

Cockerill, F. R. & CLSI. (2012). *Performance standards for antimicrobial susceptibility tests: Approved standard- eleventh edition*. *CLSI document M02-A11*. Clinical and Laboratory Standards Institute.

Department of Plant Resources. (2024). *Plants of Nepal Fact Sheet.*

Gakuubi, M. M., Wagacha, J. M., Dossaji, S. F., & Wanzala, W. (2016). Chemical composition and antibacterial activity of essential oils of *Tagetes minuta* (*Asteraceae*) against selected plant pathogenic bacteria. *International Journal of Microbiology, 2016*(1), 7352509. https://doi.org/10.1155/2016/7352509

Ghimire, S. K., Sapkota, I., Oli, B., & Parajuli, R. (2008). *Non-timber forest products of Nepal Himalaya: Database of some important species found in the mountain protected areas and surrounding regions.* WWF Nepal.

Huang, X., Xiao, Y., Köllner, T., Zhang, W., Wu, J., Wu, J., Guo, Y., & Zhang, Y. (2013). Identification and characterization of (E)-*β*-caryophyllene synthase and *α/β*-pinene synthase potentially involved in constitutive and herbivore-induced terpene formation in cotton. *Plant Physiology and Biochemistry, 73*, 302-308. https://doi.org/10.1016/j.plaphy.2013.10.017

Hulina, N. (2008). Wild marigold – *Tagetes minuta* L., new weed on the island of Hvar, and new contribution to the knowledge of its distribution in Dalmatia (Croatia). *Agriculturae Conspectus Scientificus*, *73*(1), 23-26.

Hethelyi, E., Danos, B., Tetenyi, P., Koczka, I. (1986). GC-MS analysis of the essential oils of four *Tagetes* species and the anti-microbial activity of *Tagetes minuta*. *Flavour and Fragrance Journal*, *1*,169-173.

Joshi, J., Shakya, K. S., Maharjan, S., Pradhan, S., Pradhan, R. & Ranjitkar, R. (2014). Analysis of *Tagetes minuta* L.: A potential medicinal herb. *Bulletin of the Department of Plant Resources*, *36*, 86-91.

Karimian, P., Kavoosi, G., & Amirghofran, Z. (2014a). Anti-oxidative and anti-inflammatory effects of *Tagetes minuta* essential oil in activated macrophages. *Asian Pacific Journal of Tropical Biomedicine*, *4*(3), 219-227. https://doi.org/10.1016/S2221-1691 (14)60235-5

Karimian, P., Kavoosi, G., &Amirghofran, Z. (2014b). Anti-oxidative and anti-inflammatory effects of *Tagetesminuta*essential oil in activated macrophages. *Asian Pacific Journal of Tropical Biomedicine*, *4*(3), 219-227. https://doi.org/10.1016/S2221-1691 (14)60235-5

Khakurel, B., Pradhan, R. & Lakhey, P. B. (2014). A preliminary screening of some Nepalese medicinal plants for antimicrobial activity. *Bulletin of the Department of Plant Resources*, *36*, 72-75.

Kim, B. N., Kim, N. J., Kim, M. N., Kim, Y. S., Woo, J. H., & Ryu, J. (2003). Bacteraemia due to tribe Proteeae: A review of 132 cases during a decade (1991-2000). *Scandinavian Journal of Infectious Diseases*, *35*(2), 98-103. https://doi.org/10.1080/0036554021000027015

Kunwar, R. M., Thapa-Magar, K. B., Subedi, S. C., Kutal, D. H., Baral, B., Joshi, N. R., Adhikari, B., Upadhyaya, K. S., Thapa-Magar, S., Ansari, A. S., Thapa, G. J., & Bhandari, A. R. (2023). Distribution of important medicinal plant species in Nepal under past, present, and future climatic conditions. *Ecological Indicators*, *146*, 109879. https://doi.org/10.1016/j.ecolind.2023.109879

Kyarimpa, C., Isaiah, N. O., John, D. K., Christine,

B. N., John, W., & Cliff, R. K. (2015). Evaluation of anti-oxidant properties in essential oil and solvent extracts from *Tagetes minuta*. *African Journal of Pure and Applied Chemistry*, *9*(5), 98-104. https://doi.org/10.5897/AJPAC2015.0616

Lamichhane, N., Bashyal, H. P., Thapa, A., Pant, R. R., Thapa, L. B., & Yadav, R. K. P. (2025). Naturalized exotic weed *Tagetes minuta*: A potentially invasive species in Nepal. *Korean Journal of Plant Taxonomy*, *55*, 36-43. https://doi.org/10.11110/kjpt.2025.55.1.36

Lee, E., & Anjum, F. (2024). *Staphylococcus epidermidis infection in Stat Pearls*. Stat Pearls Publishing. http://www.ncbi.nlm.nih.gov/books/NBK563240/

Lim, L. L., Chow, E., & Chan, J. C. N. (2023). Cardio-renal diseases in type 2 diabetes mellitus: Clinical trials and real-world practice. *Nature Reviews Endocrinology*, *19*(3), 151-163. https://doi.org/10.1038/s41574-022-00776-2

Mercier, B., Prost, J., & Prost, M. (2009). The essential oil of turpentine and its major volatile fraction (α- and β- pinenes): A review. *International Journal of Occupational Medicine and Environmental Health*, *22*(4). https://doi.org/10.2478/v10001-009-0032-5

Meshkatalsadat, M. H., Safaei-Ghomi, J., Moharramipour, S., & Nasseri, M. (2010). Chemical characterization of volatile components of *Tagetes minuta*: Cultivated in south west of Iran by nano scale injection. *Digest Journal of Nano-materials and Biostructures*, *5*(1), 101-106.

Oyenihi, O. R., Delgoda, R., & Matsabisa, M. G. (2021). *Tagetes minuta* leaf extracts triggered apoptosis in MCF-7 human breast cancer cell line. *South African Journal of Botany*, *137*, 359-364. https://doi.org/10.1016/j.sajb.2020.10.032

Pillai, M. K., Santi, L. I., & Mekbib, S. B. (2020). Antimicrobial activities of extracts from stem bark of *Tagetes minuta*. *Food Research*, *4*(6), 2089-2094. https://doi.org/10.26656/fr.2017.4(6).279

Pradhan, S., Paudel, H. R., Maharjan, R., & Sharma, K. (2023). Essential oils from six aromatic plants of Langtang National Park: Insights on their chemical constituents via GC-MS analysis. *Separations*, *10*(1), 52.

Rathore, S., Walia, S. & Kumar, R. (2018). Biomass and essential oil of *Tagetes minuta* influenced by pinching and harvesting stage under high precipitation conditions in the western Himalayas. *Journal of Essential Oil Research*, *30*(5), 360-368. https://doi.org/10.1080/10412905.2018.1486744

Sadia, S., Khalid, S., Qureshi, R., & Bajwa, A. A. (2013). *Tagetes minuta* L., a useful underutilized plant of family *Asteraceae*: A review. *Pakistan Journal of Weed Science Research*, *19*(2).

Sahay, R., & Patra, D. D. (2013, December 1). Identification and performance of stress-tolerant phosphate-solubilizing bacterial isolates on *Tagetes minuta* grown in sodic soil. *Soil Use and Management*, *29*(4). https://doi.org/10.1111/sum.12081

Senatore, F., Napolitano, F., Mohamed, A. H., Harris, P. J. C., Mnkeni, P. N. S. & Henderson, J. (2004). Antibacterial activity of *Tagetes minuta* L. (Asteraceae) essential oil with different chemical composition. *Flavour Fragr. J.*, *19*, 574-578

Shahzadi, I., & Shah, M. M. (2015). Acylated flavonol glycosides from *Tagetes minuta* with antibacterial activity. *Frontiers in Pharmacology*, *6*. https://doi.org/10.3389/fphar.2015.00195

Silori, G., Kushwaha, N., & Kumar, V. (2019). Essential oils from Pines: Chemistry and applications. In S. Malik (Ed.), *Essential Oil Research* (pp. 275-297). Springer Nature Switzerland AG. https://doi.org/10.1007/978-3-030-16546-8_10

Singh, V., Singh, B., & Kaul, V. K. (2003). Domestication of wild marigold (*Tagetes minuta* L.) as a potential economic crop in western Himalaya and north Indian plains. *Economic Botany*, *57*(4), 535-544.

Singh, P., Krishna, A., Kumar, V., Krishna, S., Singh, K., Gupta, M., & Singh, S. (2015). Chemistry and biology of industrial crop *Tagetes* species: A review. *Journal of Essential Oil Research*, *28*(1), 1-14. https://doi.org/10.1080/10412905.2015.1076740

Tokano, M., Tarumoto, N., Imai, K., Sakai, J., Maeda, T., Kawamura, T., Seo, K., Takahashi, K., Yamamoto, T., & Maesaki, S. (2023). Bacterial Meningitis Caused by *Bacillus subtilis* var. Natto. *Internal Medicine*, *62*(13), 1989-1993. https://doi.org/10.2169/internalmedicine.0768-22

Tong, S. Y. C., Davis, J. S., Eichenberger, E., Holland, T. L., & Fowler, V. G. (2015). *Staphylococcus aureus* Infections: Epidemiology, pathophysiology, clinical manifestations, and management. *Clinical Microbiology Reviews*, *28*(3), 603-661. https://doi.org/10.1128/CMR.00134-14

Trombetta, D., Castelli, F., Sarpietro, M. G., Venuti, V., Cristani, M., Daniele, C., Saija, A., Mazzanti, G., & Bisignano, G. (2005). Mechanisms of antibacterial action of three monoterpenes. *Antimicrobial Agents and Chemotherapy*, *49*(6), 2474-2478. https://doi.org/10.1128/AAC.49.6.2474-2478.2005

Verma, N., Aggarwal, N., & Sood, P. (2024). Exploring the phyto-chemistry and biological potential of *Tagetes minuta* L.: A comprehensive review. *South African Journal of Botany*, *168*, 175-195. https://doi.org/10.1016/j.sajb.2024.03.004

Walia, S., & Kumar, R. (2020). Wild marigold (*Tagetes minuta* L.) an important industrial aromatic crop: Liquid gold from the Himalaya. *Journal of Essential Oil Research*, *32*(5), 373-393. https://doi.org/10.1080/10412905.2020.1813211

Walia, S., Mukhia, S., Bhatt, V., Kumar, R., & Kumar, R. (2020). Variability in chemical composition and antimicrobial activity of *Tagetes minuta* L. essential oil collected from different locations of Himalaya. *Industrial Crops and Products*, *150*, 112449. https://doi.org/10.1016/j.indcrop.2020.112449

Wanzala, W., &Ogoma, S. B. (2013). Chemical composition and mosquito repellency of essential oil of *Tagetes minuta* from the southern slopes of Mount Elgon in Western Kenya. *Journal of Essential Oil Bearing Plants*, *16*(2), 216-232. https://doi.org/10.1080/0972060X.2013.793975

Journal of Plant Resources (2025) 23(1), 57-66
https://doi.org/10.3126/bdpr.v23i1.83351

Assessment of Soil Physicochemical Characteristics and NDVI in Response to Forest Vegetation Types of Godawari Kunda Community Forest, Lalitpur, Nepal

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Abstract

This study focused on the relationship between soil physicochemical characteristics and Normalized Difference Vegetation Index (NDVI) in response to forest vegetation types in Godawari Kunda community forest, Lalitpur, Nepal. It aimed to provide valuable interactions of forest ecosystems by investigating how different vegetation types influence soil properties, nutrient cycling, and NDVI. The objectives included assessing soil physicochemical characteristics in response to forest vegetation types and examining NDVI variation across forest types. A stratified random sampling method resulted in 45 plots (10 × 10 m² each) across three altitudinal ranges: *Schima-Castanopsis* forest at the base (1400–1600 m), Mixed Broadleaf forest (1601–1800 m), and Temperate Broadleaf forest (1801–2000 m). The results showed the highest NDVI value (0.75) in the Temperate Broadleaf forest (1801–2000 m), while the lowest NDVI value (0.36) was observed in the *Schima-Castanopsis* forest (1400–1600 m), corresponding to tree density variations. Significant variations in physical and chemical soil characteristics were also evident. Positive correlation was observed between soil organic carbon, available nitrogen and soil moisture with forest vegetation types and NDVI. On the other hand, soil pH and potassium levels showed negative correlations with NDVI and vegetation types. These findings offer valuable data for land-use planning, soil carbon stock assessment, and climate change studies, emphasizing the importance of soil-vegetation relationships for effective ecosystem management, biodiversity conservation, and sustainable land use practices.

Keywords: Mixed Broadleaf forest, NDVI, *Schima-Castanopsis* forest, Soil characteristics, Temperate broadleaf forest

Introduction

Forest ecosystems are constantly changing settings where the relationships between plant life and soil characteristics are crucial for maintaining ecosystem functions and productivity. These relationships are additionally affected by elevation, which determines climate patterns, the distribution of vegetation, and the formation of soil (Banday et al., 2019; Imran et al., 2021). The physicochemical properties of soil, including pH, texture, organic carbon, nitrogen, and potassium, are crucial for the growth of plants and the stability of ecosystems. Changes in these soil characteristics can affect species diversity, nutrient cycling, and the overall health of forests (Dahal et al., 2018; Schoonover & Crim, 2015). Remote sensing technologies, especially the Normalized Difference Vegetation Index (NDVI), are essential tools for evaluating vegetation cover, biomass, and photosynthetic processes. NDVI measures spectral reflectance in the red and near-infrared wavelengths to assess the health of vegetation, with higher NDVI readings reflecting lush, thriving vegetation and lower readings indicating sparse or degraded cover (Tucker, 1979; Yacouba et al., 2010). In mountain ecosystems, variations in altitude lead to diverse types of vegetation, which often correlate with alterations in soil characteristics and NDVI values (Banday et al., 2019; Imran et al., 2021). Analyzing the variations of NDVI among various forest types and altitude levels can offer valuable information regarding vegetation productivity, land cover changes, and overall ecosystem health.

The Godawari Kunda community forest is located in the Godawari Municipality within the Lalitpur district, positioned on the southern inclines of Phulchowki Hill in central Nepal (27°352 07.473 N, 85°242 05.183 E). This area ranges in elevation from 1400 to 2000 meters above sea level (asl) and serves as a transitional zone between subtropical and temperate climate regions (Kharbuja & Rajbhandary, 2022). The forest consists of three primary types of vegetation-*Schima*-*Castanopsis* forest in the lower regions, Mixed Broadleaf forest in the middle regions, and Temperate Broadleaf forest in the upper regions. Godawari and Phulchowki are noted as important biodiversity hotspots in Nepal, hosting a variety of plant and animal life, including species that are endemic and at risk. Although the ecological and conservation importance is recognized, there are few integrated studies that investigate the connection between soil physicochemical properties, types of forest vegetation, and remotely sensed NDVI data in this region. However, this research intends to evaluate how different types of forest vegetation affect the physicochemical properties of soil and NDVI in the Godawari Kunda community forest located in Nepal. The specific objectives include to assess the variation in NDVI, evaluating the soil physical and chemical characteristics in response to forest vegetation types of Godawari Kunda community forest.

Material and Methods

Study area

The study has been carried out, in Godawari Kunda community forest which is situated in Godawari Municipality, Phulchowki hill, Lalitpur district of Bagmati zone, Nepal (27°35’ 07.47"N, 85°24’05.18"E) (Figure 1). Phulchoki Hill is located in the area where the subtropical and temperate climates meet in the southern Kathmandu Valley (Kharbuja & Rajbhandary, 2022). The altitude of Godawari Kunda community forest hill ranges from 1400 to 2006 m asl.

The study site is located inside the territory of Phulchowki Hill and three different types of evergreen broad-leaved forests make up Godawari Kunda community forest vegetation[a *Schima*-*Castanopsis* forest at the base (1400 m to 1600 m), Mixed broadleaf forest (1600 m to 1800 m), and Temperate Broadleaf forest (1800-2000 m)].

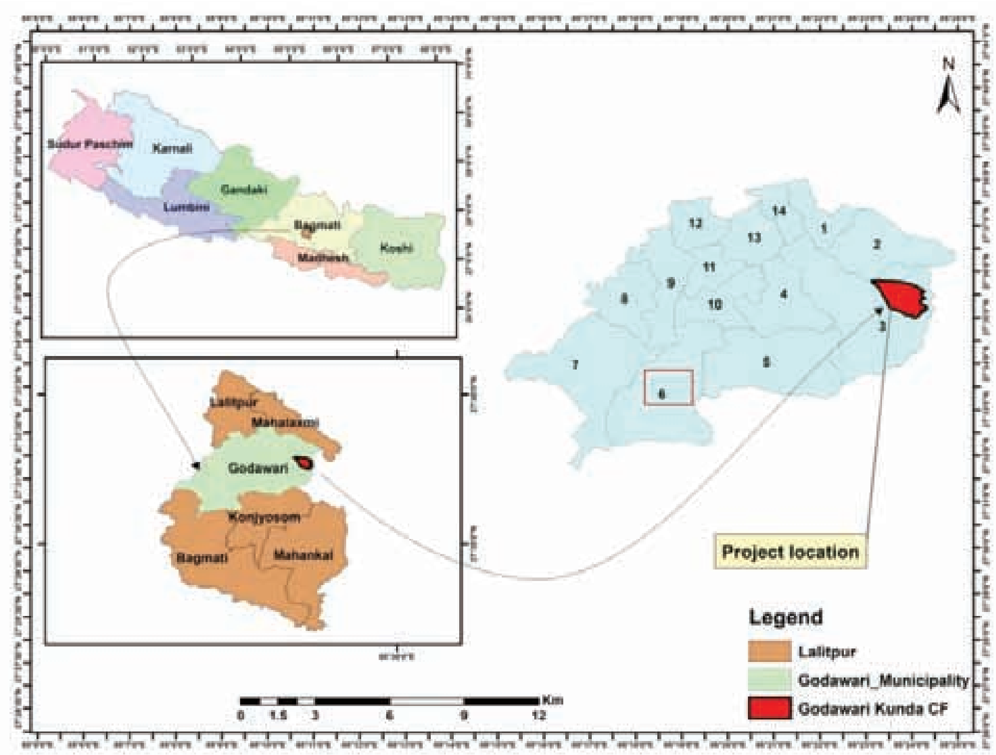

Figure 1: Study site

Field visits and soil sample collection

A stratified random sampling approach was employed to investigate the physicochemical properties of soil across three types of forest vegetation: *Schima-Castanopsis* (1400-1600 m), Mixed Broadleaf (1601-1800 m), and Temperate Broadleaf (1801-2000 m). For each stratum, 15 plots measuring 10 × 10 m were established through a randomized block design, culminating in a total of 45 plots. Soil samples were taken from two different depths (0-15 cm and 15-30 cm) with the help of a soil borer and shovel. The sampling was conducted in triplicate for every depth and forest type, leading to a total of 90 composite soil samples. Each sample was placed in a labeled polythene bag, and the fresh weight was measured on-site using a portable digital scale. GPS coordinates and aspect details were noted for all plots.

Laboratory analysis of soil parameters

Soil pH was assessed with a digital pH meter in a 1:2.5 mixture of soil and water (Jackson, 1973). The soil texture, including sand, silt, and clay, was analyzed using the hydrometer method (Bouyoucos, 1962). The organic carbon content was determined through the Walkley-Black dichromate oxidation procedure (Walkley & Black, 1934). Total nitrogen levels were quantified using the Kjeldahl digestion technique (Bremner & Mulvaney, 1982), while available potassium was measured through flame photometry after extraction with ammonium acetate (Jackson, 1973). Moisture content was calculated gravimetrically by drying fresh samples at 105°C for at a duration of 24 hours.

Laboratory tests for organic matter and carbon were performed at the SchEMS College Environmental Laboratory, while the analyses for nitrogen and potassium were conducted at the Laboratory of Department of Environmental Science, Tribhuvan University, Kathmandu.

NDVI data acquisition and processing

NDVI values were obtained from Sentinel-2A multispectral satellite images with a resolution of 10 m, accessed through the Copernicus Open Access Hub for the years 2017 and 2022. The pre-processing phase involved atmospheric correction via the Sen2Cor plugin in SNAP software. NDVI was computed using the conventional formula:

$$NDVI = \frac{(\mathrm{NIR} - \mathrm{RED})}{(\mathrm{NIR} + \mathrm{RED})}$$

Where, NIR and RED refer to the near-infrared (Band 8) and red (Band 4) bands, respectively. NDVI maps and classification were generated using QGIS 3.10

Data analysis

Statistical analyses were conducted using SPSS (version 20) and Microsoft Excel. Descriptive statistics were used to summarize the data. To assess differences among forest types, one-way ANOVA was performed for normally distributed variables, with Tukey's B post-hoc test for pair wise comparisons. For non-normally distributed data, the Kruskal-Wallis H test was used. Significant differences were considered at $p < 0.05$. Correlation analysis (Pearson's r) was conducted to examine relationships between NDVI and soil parameters. The significance of differences in soil parameters across depth and forest type was further interpreted based on F-values (ANOVA) and χ^2 values (Kruskal-Wallis), as detailed in the results section.

Results and Discussion

The physicochemical properties of soil showed considerable variation among the three types of forest vegetation and different soil depths in the Godawari Kunda community forest. The analyzed parameters included soil texture, moisture levels, pH, organic carbon (OC), total nitrogen (N), available potassium (K), phosphorus (P), and NDVI. Statistical tests (ANOVA, Kruskal-Wallis) indicated significant differences in most parameters across the forest types ($p < 0.05$), especially in NDVI, soil moisture, organic carbon, nitrogen, and phosphorus.

NDVI variation and tree density

NDVI values were highest in the Temperate Broadleaf forest (mean NDVI = 0.75), followed by the Mixed Broadleaf forest (0.52), and lowest in the *Schima-Castanopsis* forest (0.36) (Figure 2). One-way ANOVA revealed a statistically significant difference in NDVI values among the three forest types (F(2,42), $p < 0.001$), indicating that vegetation greenness significantly varies with forest type.

The higher NDVI values observed in upper elevation forests correspond with greater tree density and enhanced soil fertility. This pattern supports previous research by Banday et al. (2019) and Imtimongla et al.(2021), which reported increased canopy vigor and vegetation density at higher elevations. Pearson correlation analysis further demonstrated that NDVI is significantly and positively correlated with organic carbon (r = 0.79), nitrogen (r = 0.74), and soil moisture (r = 0.68), while it is negatively correlated with soil pH (r = -0.62) and potassium (r = -0.57), all of which were statistically significant ($p < 0.01$).

In terms of forest structure, the highest tree density was recorded in the Temperate Broadleaf forest located at elevations between 1801-2000 m, where tree height ranged from 10.1 to 20 meters. This forest type contained 340 trees per hectare, significantly higher than the densities observed in other forest types ($p < 0.05$, Tukey HSD post-hoc test). Conversely, the *Schima-Castanopsis* forest, located at lower elevations (1400–1600 m), exhibited the lowest tree density, with only 100 trees per hectare and dominant tree heights ranging from 20.1 to 30 meters (Figure 3).

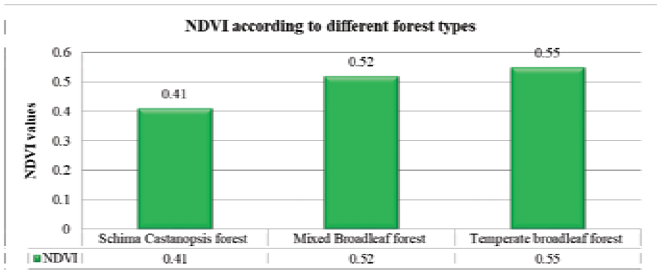

Figure 2: NDVI values according to different forest types

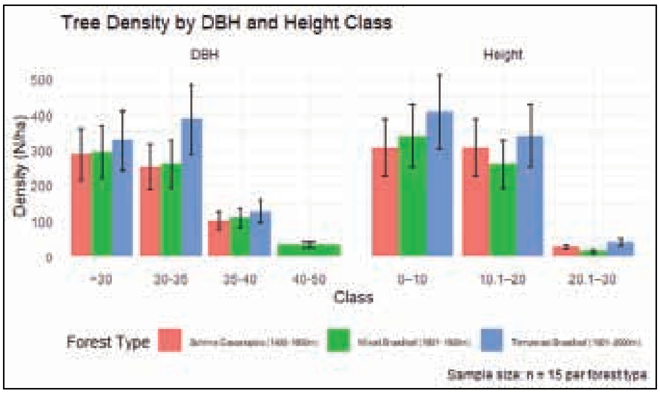

Figure 3: Density of the forest according to forest vegetation types

These findings indicate a statistically significant variation in tree density across forest types, which is consistent with the NDVI distribution observed

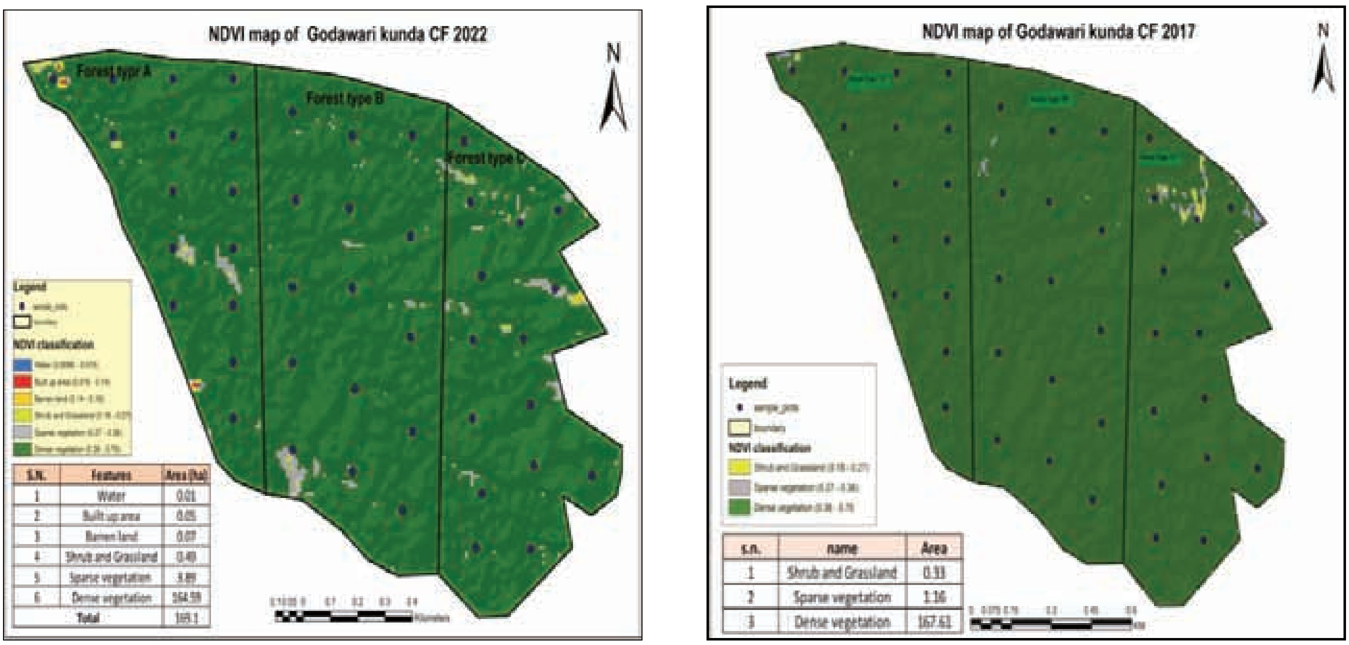

Figure 4: NDVI maps of Godawari Kunda community forest in year 2017 and 2022

in Sentinel-2A imagery (Figure 4). The strong association between NDVI and tree density suggests that NDVI can be a reliable proxy for assessing vegetation structure and forest health in the study area.

Physical properties

Soil texture: Soil texture displayed slight yet consistent changes along elevation gradients. Soils of the *Schima-Castanopsis* forest (1400-1600 m) possessed the greatest sand content (94.33%) along with the least clay content (3.73% at 0-15 cm), reflecting a coarser texture with reduced capacity for holding water. Conversely, the Temperate Broadleaf forest (1801-2000 m) had the highest clay content (5.2% at 15-30 cm) and the lowest sand content (92.2%), which enhances its ability to retain moisture (Table 1, Figure 5).

Moisture: Moisture levels rose notably with elevation at both soil depths. In the upper layer (0-15 cm), the average soil moisture recorded was 22.65% in the *Schima-Castanopsis* forest, 42.20% in the Mixed Broadleaf forest, and 58.15% in the Temperate Broadleaf forest. For the deeper layer (15-30 cm), the corresponding moisture percentages were 17.74%, 36.72%, and 52.12%, respectively (Table 2, Figure 6). Statistical evaluations indicated significant differences in moisture content among the various forest types ($F(2,42) = 89.854$ for 0-15 cm; $F(2,42) = 91.704$ for 15-30 cm; $p < 0.001$). The elevated moisture levels in higher elevation forests foster microbial activity and the breakdown of organic matter, which facilitates nutrient cycling and accumulation (Imtimongla et al., 2021).

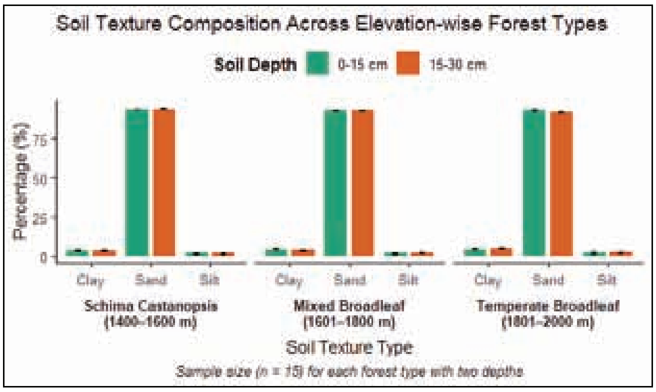

Figure 5: Clay, slit and sand content in soil response to forest vegetation types

Table 1: Descriptive statistics of clay, slit and sand content in soil response to forest vegetation types

Forest Types	Soil Texture	Descriptive Statistics							
		Soil depth (0-15 cm)				Soil depth (15-30cm)			
		Mean±SE	SD	Min.	Max	Mean±SE	SD	Min.	Max
Schima-Castanopsis forest (1400-1600m)	Silt	1.93±0.25	0.98	0.5	3	2±0.25	0.98	0	4
	Clay	3.73±0.18	0.73	2.3	4.3	3.67±0.26	1.008	2.3	5.3
	Sand	94.33±0.27	1.04	92.7	96.7	94.33±0.38	1.47	92.7	97.7
Mixed Broadleaf forest (1601-1800 m)	Silt	2±0.29	1.15	1	4.5	2.53±0.45	1.74	0	7
	Clay	4.5±0.29	1.105	2.6	6.1	4.167±0.35	1.35	2.1	6.6
	Sand	93.5±0.39	1.54	89.4	95.4	93.3±0.54	2.09	89.4	97.4
Temperate Broadleaf forest (1801-2000 m)	Silt	2.167±0.52	2.023	0	6	2.6±0.55	2.13	0.5	8
	Clay	4.5±0.20	0.78	3.1	5.6	5.2±0.19	0.74	4.1	6.6
	Sand	93.33±0.64	2.49	88.4	96.4	92.2±0.68	2.62	85.4	95.4

Table 2: Descriptive statistics of moisture content (%) in soil in response to forest vegetation types

Forest Types	Descriptive Statistics							
	Soil depth (0-15 cm)				Soil depth (15-30 cm)			
	Mean±SE	SD	Min	Max	Mean±SE	SD	Min	Max
Schima-Castanopsis forest (1400-1600 m)	22.65±2.02	7.82	11	33.33	17.74±1.79	6.73	8.1	29.03
Mixed Broadleaf forest (1601-1800 m)	42.20±1.9	7.39	29.03	53.85	36.72±1.86	7.21	25	48.14
Temperate Broadleaf forest (1801-2000 m)	58.15±1.69	6.53	48.14	66.67	52.12±1.56	6.04	42.86	60

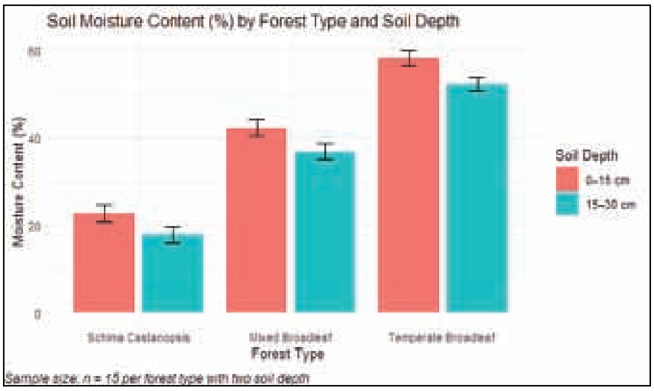

Figure 6: Moisture content (%) in soil in response to forest vegetation types

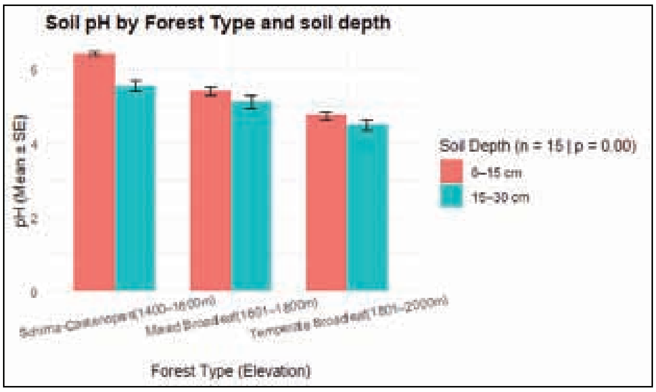

Figure 7: Difference in pH in different forest types

Chemical properties

pH: Soil pH was found to decline as altitude increased, with values ranging from 6.7 in the *Schima-Castanopsis* forest to 4.49 in the Temperate Broadleaf forest (Table 3, Figure7). Significant differences were identified among different forest types and soil depths through ANOVA and Kruskal-Wallis tests (F (2,42) = 11.115 at 0-15 cm, $\chi 2(2)$ = 18.219 at 15-30 cm; $p < 0.001$). The reduction in pH is linked to the accumulation of organic matter and leaching processes occurring at higher elevations (Tellen& Yerima, 2018).

Organic carbon, nitrogen, potassium and phosphorous: The content of organic carbon and nitrogen increased with altitude. The highest level of OC was found in the Temperate Broadleaf forest (3.99% at 0-15 cm), with notable differences observed among various forest types (F(2,42) = 15.580 at 0-15 cm; F(2,42) = 26.141 at 15-30 cm; $p < 0.001$). A similar pattern was observed for nitrogen content, which peaked in the upper forest (F(2,42) = 15.580 at 0-15 cm; F(2,42) = 26.141 at 15-30 cm; $p < 0.001$). These findings are consistent with those of Banday et al.(2019) and Shelukindo et al.(2014), who found elevated levels of SOC and N in cooler, less disturbed forests that have abundant litter input.

Table 3: Descriptive statistics of chemical properties in response to forest vegetation types

Chemical Properties	Forest Types*	Descriptive Statistics							
		Soil depth (0-15 cm)				Soil depth (15-30cm)			
		Mean±SE	SD	Min.	Max	Mean±SE	SD	Min.	Max
pH	A	6.4±0.044	0.173	6.2	6.7	5.54±0.153	0.59	4.3	6.3
	B	5.4±0.128	0.49	4.4	6.2	5.11±0.176	0.68	3.6	6
	C	4.73±0.106	0.412	4.1	5.3	4.49±0.146	0.56	3.7	5.3
OC	A	2.52±0.24	0.92	0.77	4.27	1.75±0.19	0.74	0.39	3.11
	B	2.63±0.24	0.93	0.78	3.69	1.98±0.22	0.84	0.39	3.30
	C	3.99±0.13	0.49	3.11	4.66	3.54±0.16	0.61	2.72	4.66
N	A	0.43±0.04	0.16	0.134	0.74	0.302±0.03	0.13	0.067	0.536
	B	0.45±0.04	0.16	0.134	0.64	0.34±0.04	0.14	0.067	0.57
	C	0.69±0.022	0.08	0.536	0.804	0.609± 0.027	0.11	0.469	0.804
P	A	14.56±0.52	2.03	12.11	17.85	13.50±0.59	2.28	10.21	17.43
	B	18.73±1.19	4.59	11.39	26.71	16.87±1.12	4.36	10.17	26.21
	C	21.21±0.91	3.53	16.67	26.16	20.23±0.92	3.57	15.66	25.07
K	A	411.98±74.51	288.57	217.73	1306.37	302.67±19.99	77.43	193.54	502.66
	B	323.81±26.91	104.22	209.66	545.66	308.76±23.63	91.52	215.04	532.22
	C	328.65± 26.94	104.33	137.088	567.17	298.55±21.36	82.71	139.78	405.89

*Note: *Schima-Castanopsis* forest (1400-1600m), Mixed Broadleaf forest (1601-1800 m), Temperate Broadleaf forest (1801-2000 m) are referred as Forest type 'A', 'B' and 'C'

The levels of available potassium did not exhibit notable differences across various forest types (Kruskal-Wallis $\chi2(2) = 0.418$ at 0-15 cm; $\chi2(2) = 0.000$ at 15-30 cm; $p > 0.05$). This lack of variation may be attributed to a consistent parent material or a stable mineral composition (Imran et al., 2021). On the other hand, phosphorus exhibited significant variability among forest types ($F(2,42) = 13.442$ at 0-15 cm; $F(2,42) = 13.752$ at 15-30 cm; $p < 0.001$), indicating its susceptibility to biological cycling and the effects of forest management strategies (Kidanemariam et al., 2012).

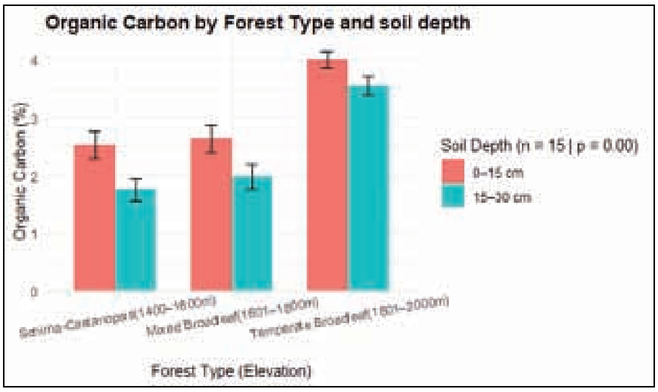

Figure 8: Difference in organic carbon in different forest types

Within the temperate broadleaf forest (1801-2000 m), the organic carbon content reached its peak at 4.66% in the 0-15 cm and 15-30 cm depth, while the lowest content of 0.39% was observed in the 15-30 cm depth of the Mixed Broadleaf forest (1601-1800 m) and *Schima-Castanopsis* forest (1400-1600 m) (Table 3, Figure 8).With rising altitudinal ranges, the organic carbon content rose (Banday et al., 2019) given reason that the persistent accumulation of leaf litter and the slower decomposition rate caused by the lower temperature can be attributed to the larger levels of organic carbon in higher altitudes. Higher elevations will have more organic carbon content because slower breakdown can result in less mineralization and less erosion of organic carbon. Also, seen in the result of research done by Shelukindo et al. (2014) that compared to the relatively lower elevation, the relatively higher elevation conditions encouraged significantly more SOC buildup.

In Godawari Kunda community forest, it has been observed that potassium exhibits a negative correlation with various types of forest vegetation. Among the different forest types, the *Schima-Castanopsis* forest (1400-1600 m) showed the highest potassium level at a soil depth of 0-15 cm, measuring 1306.37 kg/ha. On the other hand, the Temperate Broadleaf forest (1801-2000m) had the lowest potassium content, with a measurement of 137.088 kg/ha at the 0-15 cm soil depth (Table 3, Figure 9). It may be because of altitude influencing the type and composition of rocks and minerals in an area. Some rocks contain potassium-bearing minerals. However, at higher altitudes, the types of rocks present may be less likely to contain significant amounts of potassium-rich minerals. This limited availability of potassium-bearing minerals can contribute to lower potassium levels in the soil (Imran et al., 2021).

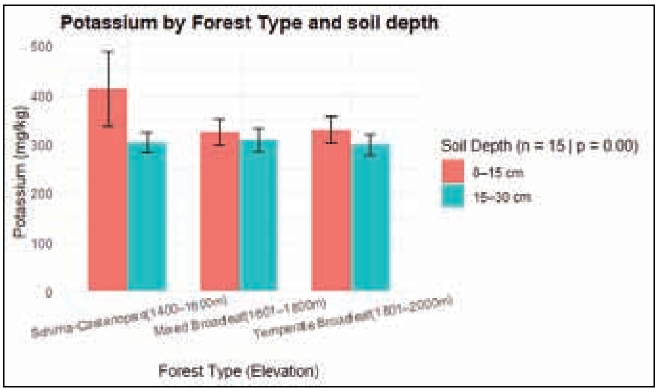

Figure 9: Difference in content of potassium in different forest types

The results of the nitrogen content analysis in the soil revealed varying levels among different forest types at different altitudes. The highest nitrogen content, measuring 0.804%, was observed in the Temperate Broadleaf forest located between 1801-2000 meters above sea level. On the other hand, the lowest nitrogen content, measuring 0.067%, was found in both the *Schima-Castanopsis* forest (1400-1600m) and Mixed Broadleaf forest (1601-1800m) (Table 3, Figure 10). These measurements were taken at a soil depth of 0-15 and 15-30 centimeters. It was found that the amount of nitrogen content increased with increase in response to forest vegetation. Therefore, with rising NDVIs, the amount of available N, P, K, Ca, and S increased (Banday et al., 2019). Same result was observed by Imtimongla et al.(2021), high SOM may be the cause of the soil's high total N concentration with elevation.

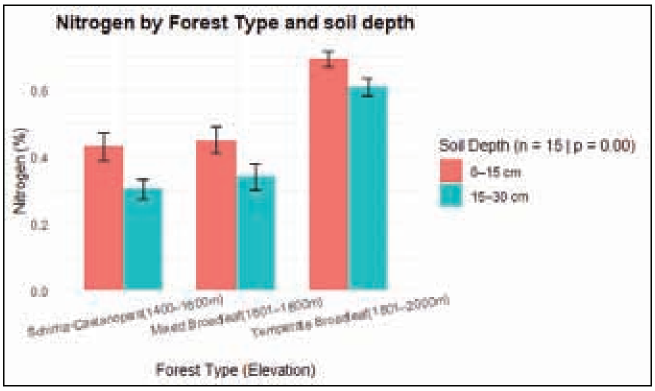

Figure 10: Difference in nitrogen content in different forest types

The analysis of available phosphorus in the soil across different forest types and altitudes revealed a clear variation. The highest phosphorus content was recorded in Temperate Broadleaf forest with a mean value of 21.21 ± 0.91 mg/kg at 0-15 cm depth and 20.23 ± 0.92 mg/kg at 15-30 cm depth. In contrast, the lowest phosphorus concentration was found in *Schima-Castanopsis* forest, measuring 14.56 ± 0.52 mg/kg at 0-15 cm and 13.50 ± 0.59 mg/kg at 15-30 cm. (Mixed Broadleaf forest showed intermediate phosphorus levels (Table 3, Figure 11). The trend of increasing phosphorus with elevation and forest type can be attributed to higher organic matter accumulation and nutrient cycling efficiency in upper elevation forests. These results are consistent with the findings of Banday et al. (2019), who

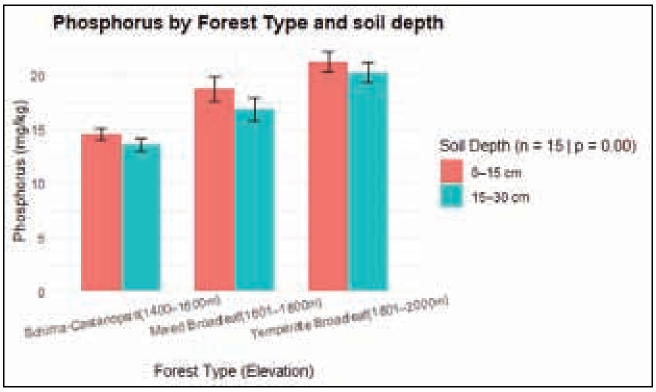

Figure 11: Difference in phosphorous content in different forest types

reported that available phosphorus content tends to increase with higher NDVI and forest productivity. Similarly, Imtimongla et al.(2021) observed elevated phosphorus levels in soils with higher soil organic matter (SOM) at greater altitudes, suggesting improved mineralization and nutrient retention capacity. The current findings support the notion that phosphorus availability is closely linked to vegetation type and altitudinal gradients.

Conclusion

This research reveals that different types of forest vegetation and variations in altitude significantly affect the physicochemical properties of soil and the health of vegetation, as measured by NDVI, within the Godawari Kunda community forest. The Temperate Broadleaf forest, located at the highest altitude, displayed notably elevated levels of NDVI, soil organic carbon, total nitrogen, and moisture, along with reduced pH levels in comparison to forests at lower elevations. These observed trends indicate a greater accumulation of nutrients, increased vegetation density, and enhanced ecological interactions in high-altitude forest systems that are less disturbed.

Statistical evaluations (ANOVA, Kruskal-Wallis, and correlation) demonstrated that the majority of soil characteristics varied considerably across different forest types, with phosphorus exhibiting significant variation while potassium remained relatively unchanged. NDVI showed a strong positive correlation with essential fertility indicators (OC, N & moisture), reinforcing its usefulness as a remote sensing tool for evaluating forest health and soil quality. The combination of satellite-derived NDVI and field-based soil analyses creates a strong framework for monitoring forest ecosystems, especially within mountainous and community-managed landscapes.

By establishing foundational connections between vegetation type, altitude, soil fertility, and NDVI, this study provides valuable insights for sustainable forest management, land-use planning, and ecosystem monitoring in the mid-hill regions of Nepal. It also emphasizes the importance of merging field ecology with geospatial methodologies for assessments at the landscape level in regions rich in biodiversity but lacking extensive data.

Author Contributions

B Timilsina & P K Regmi have contributed equally to bring the manuscript in this form and R A Mandal designed the research.

Acknowledgements

We would like to give our gratitude respect and appreciation to all the people who have helped and given us guidance during this research. A special gratitude goes to respected Late Dr. Ram Asheshwar Mandal for his regular guidance, supervision and encouragement during my research work. I would like to thank my Principal Mr. Ajay B Mathema, Academic coordinator, Mr. Praveen Kumar Regmi, and all SchEMS family for valuable advice and support throughout the research period. In particular, my special thanks go to Ms. Durga Pokharel, Mr. Ram Puri two community forests user groups member of study area who shared information and helped me throughout my research work. Also, thanks goes to Lab Teacher Mr. Ramesh Basnet Sir for his guidance and full support throughout our project lab report.

References

Banday, M., Bhardwaj, D. R., & Pala, N. A. (2019). Influence of forest type, altitude and NDVI on soil properties in forests of North Western Himalaya, India. *Acta Ecologica Sinica*, *39*(1), 50-55. https://doi.org/10.1016/j.chnaes.2018.06.001

Bouyoucos, G. J. (1962). Hydrometer method improved for making particle size analyses of soils 1. *Agronomy Journal*, *54*(5), 464-465. https://doi.org/10.2134/agronj1962.00021962005400050028x

Dahal, J., Chidi, C. L., Mandal, U. K., Karki, J., Khanal, N. R., & Pantha, R. H. (2018). Physico-chemical properties of soil in Jita and Taksar Area of Lamjung district, Nepal. *Geographical Journal of Nepal*, *11*, 45-62. https://doi.org/10.3126/gjn.v11i0.19548

Imran, Amanullah, Naveed, S., Khan, I., Sajid, M., Mahmood, T., Hussain, I., Ilyas, M., Ali, I., Ullah, S., Kamal, A., Altawaha, A. R., Al-Tawaha, A. R., Thangadurai, D., Sangeetha, J., Rauf, A., Saranraj, P., Al-Sultan, W., Al-Taey, D. K. A., … Hastang. (2021). Relationship of soil physico chemical properties with elevation and geographical directions. *IOP Conference Series: Earth and Environmental Science*, *788*(1). https://doi.org/10.1088/1755-1315/788/1/012172

Imtimongla, Ariina, M. S., & Phucho, S. D. & T. (2021). Properties of soil in relation to altitude. *Just Agriculture*, *1*(12), 1-13.

Kharbuja, A., & Rajbhandary, S. (2022). Grass flora along altitudinal gradient of the Phulchoki Hill, Central Nepal. *Journal of Plant Resources*, *20*(1), 52-63. https://doi.org/10.3126/bdpr.v20i01.56579

Kidanemariam, A., Gebrekidan, H., Mamo, T., & Kibret, K. (2012). Impact of altitude and land use type on some physical and chemical properties of acidic soils in Tsegede Highlands, Northern Ethiopia. *Open Journal of Soil Science*, *02*(03), 223-233. https://doi.org/10.4236/ojss.2012.23027

Schoonover, J. E., & Crim, J. F. (2015). An introduction to soil concepts and the role of soils in watershed management. *Journal of Contemporary Water Research & Education*, *154*(1), 21-47. https://doi.org/10.1111/j.1936-704x.2015.03186.x

Shelukindo, H. B., Semu, Msanya, Singh, & Munishi, P. K. T. (2014). Soil organic carbon stocks in the dominant soils of the Miombo woodland ecosystem of Kitonga Forest Reserve, Iringa, Tanzania. *International Journal of Agricultural Policy and Research*, *2*(4), 167-177. http://www.journalissues.org/ijapr/

Tellen, V. A., & Yerima, B. P. K. (2018). Effects of land use change on soil physicochemical properties in selected areas in the North West region of Cameroon. *Environmental Systems Research*, *7*(1). https://doi.org/10.1186/s40068-018-0106-0

Tucker, C. J. (1979). Red and photographic infrared linear combinations for monitoring vegetation. *Remote Sensing of Environment*, *8*(2), 127-150. https://doi.org/10.1016/0034-4257(79)90013-0

Walkley, A., & Black, I. A. (1934). An examination of the degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. *Soil Science*, *37*(1), 29-38. https://doi.org/10.1097/00010694-193401000-00003

Yacouba, D., Guangdao, H., & Xingping, W. (2010). Assessment of land use cover changes using Ndvi And Dem In Puer And Simao Counties , Yunnan Province , China. *Report and Opinion*, *2*(9), 7-16.

Journal of Plant Resources (2025) **23**(1), 67-79
https://doi.org/10.3126/bdpr.v23i1.83355

Utilization Status of Wetland Flora by Indigenous Peoples and Local Communities in Major Lakes of Pokhara Valley, Nepal

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Abstract

This study aims at exploring the utilization and conservation status of wetland flora by indigenous peoples and local communities by documenting the use patterns based on their indigenous and local knowledge in the three major lakes - Phewa, Begnas and Rupa in Pokhara valley, Nepal. It assesses the species distribution and ecosystem health of both aquatic and terrestrial biodiversity and ecosystem services generated by the plant species found in the lake basins of these lakes. The focus of the study is on assessing biological status of and trend in the use practice of these floral species found in and around the lakes by the communities based on their diverse and traditional practices. Three methodological tools: Plant Collection and Herbarium Analysis, Key Informant Interview (KII) and Focus Group Discussion (FGD) were applied for collection and analysis of data. The collected herbaria were cross-checked with the herbarium specimens available at the Tribhuvan University Central Herbarium (TUCH). A total of 81 wetland-based plant species of 72 genus and 42 families were recorded. Out of which 39 plants species were found being used for medicinal purposes, while 6 plant species was used for food, 4 plant species used for fodder purposes and 3 plant species used for both medicine and food purposes and only one plant species used for firewood purpose in the three-lake basin areas. Change in status of availability of plants over the years was studied with the help of the knowledge and insights of indigenous peoples and local communities. The study found out that around 18 plant species are in declining form and 6 species are in increasing form while 57 species are in stable form in and around the lake basin areas.

Keywords: Aquatic and terrestrial biodiversity, Ecosystem-based adaptation, Indigenous and local knowledge, Wetland ecosystems

Introduction

The socio-economic and ecological importance of the lake ecosystems of Pokhara Valley, particularly Phewa, Begnas, and Rupa, is significant for conserving aquatic biodiversity and supporting lake basin communities, especially in the context of climate change (https://www.ramsar.org/). These wetlands host diverse floral species providing essential ecosystem goods and services (EGS) that sustain the livelihoods of Indigenous Peoples and Local Communities (IPLCs). In Nepal, over 17% of the population is dependent on wetland resources for their livelihoods (K.C. et al., 2014). The conservation of the lake biodiversity relies heavily on preserving Indigenous and Local Knowledge (ILK), which has gained recognition for its vital role in natural resource management, particularly in countries like Nepal (Brokensha et al., 1980; Ministry of Environment, Science and Technology [MoEST],2015).There are 21 ethnic communities in Nepal that use ILK to sustainably manage natural resources (National Lake Conservation Development Committee [NLCDC], 2021; Ministry of Forest and Soil Conservation [MoFSC], 2006).

Critical role of indigenous and local knowledge in conservation

Indigenous Peoples and Local Communities (IPLCs) have relied on wetland resources for generations as an integral part of their livelihoods. However, in recent years, excessive human pressure and weak resource governance by government and community-based agencies have led to the degradation of these ecosystems, disrupting the age-old relationship between IPLCs and wetlands. Communities extract resources such as fodder, fuelwood, fiber, and medicinal plants using both modern methods and ILK. ILK is traditionally linked to conservation, sustainable harvesting, and cultural or religious practices. Scientific knowledge is also applied, particularly in fishing, green felling, and agricultural expansion. The loss of ILK can lead to ecological degradation as ethical and sustainable practices are replaced by short-term economic interests (Karki &Adhikari, 2015; Loh & Harmon, 2014). When species used in traditional practices are forgotten, they may lose their cultural significance and protection, posing risks to both biodiversity and traditional knowledge systems (Brosi et al., 2007; Karki &Adhikari, 2015). Documenting ILK is, therefore, vital not onlyfor cultural preservation but also for promoting sustainable development (Mercon et al., 2019).

In Nepal, ILK systems have been ignored in the past but are gradually being recognized and valued. Farmers in the Tarai and Midhills regions have developed different agro-forestry models and adaptation practices to cope with recurring floods, droughts, landslides and high rates of soil erosion. This diverse knowledge system demonstrates that indigenous and local knowledge and practices are at the core of the community resilience building process due to their inherent strength in anticipating and preparing for disasters before they occur (International Centre for Integrated Mountain Development [ICIMOD], 2007; Karki & Adhikari, 2015; MoEST, 2015). Indigenous and local knowledge also contributes to building social capital, which is essential for securing and enhancing livelihood opportunities (Berkes et al., 2000). Although there are still gaps in the systematic use of ILKP, several agencies and researchers (Egeru, 2012; Karki et al., 2017) have made efforts to develop uniform approaches, participatory processes and mechanisms for working with indigenous communities and promoting local knowledge systems.

However, the loss and degradation of ILK in the context of wetland conservation in the Pokhara Valley in recent years have hampered the planning and implementation of ecosystem-based adaptation and resilience building effort (Dixit et al., 2015). Such indigenous and local knowledge pools that are handed over through generations, are rich in both social and technical dimensions and are highly applicable to ecosystem based adaptation strategies (Karki et al., 2017). The availability of diverse and rich ILK and skill, has made local communities' people in the region particularly knowledgeable and skillful in the use and application of plant biodiversity based conservation values and practices. Many traditional systems of forest and water management systems use ILK, which continues to guide sustainable resource use. Despite its potential, this valuable knowledge pool remains unexplored, poorly archived and largely underutilized in the current ecosystem based adaptation (EbA) strategies and action plans are concerned (MoEST, 2015).

Very few research works have focused on the growing threats to wetland ecosystem and their associated floral biodiversity. Invasive Alien Plant Species (IAPS) is now one of the most significant drivers of plant population declines and species extinctions, particularly in island and fragile ecosystems worldwide (Donlan et al., 2003; Reaser et al., 2007). There are 166 alien plant species in Nepal, of which, herbs account for approximately 76%, followed by shrubs 16%, climbers 6% and trees 2% (Siwakoti, 2012).These invasive species pose a serious threat to native plant communities, especially in wetland habitats, where competition for space and nutrients is intense.

Despite such threats, Nepal is known for its rich biodiversity and cultural diversity. According to the 2021 Census, there are 142 caste and ethnic groups in the country (Guragain, 2024). Many of these groups maintain a deep connection with their surrounding ecosystems through ILK). However, documentation of wetland species and associated ILK remains limited, largely due to the lack of understanding about their inter relationship. This is particularly concerning given that an estimated 101 ethnic groups living in Nepal rely directly or indirectly on local biodiversity and ecosystem services. The largest ethnic group is Brahmin (27%), followed by Gurung (16%) and Chhetri (14%), while 89 ethnic groups comprise less than 1% of the total population (Central Bureau of Statistics [CBS], 2014). In the study area, around 21 Indigenous Peoples and Local Communities (IPLCs) are found. Agriculture is the major source of livelihood, supporting around 72.4% of population draw their livelihood in the region. Agriculture is supplemented by animal husbandry and fish farming in the rural and lake areas (https://lgcdp.gov.np/gandakiinfo).

This study was undertaken to document indigenous and local knowledge as well as assess the status and trends, and practices concerning the availability, utilization and conservation of wetland-based plant resources in three major lakes (Phewa, Begnas, and Rupa) of the Pokhara Valley. The finding are expected to inform efforts to conserve plant resources and to prevent the further loss of biodiversity and associated ILK necessary for the protection and sustainable use and management of aquatic and lake basin area-based plant resources.

Materials and Methods

Study area

The study was carried out in and around the three major lakes (Phewa, Rupa and Begnas) of Pokhara Valley, Gandaki province, Central Nepal (Figure 1). The climate of the study area is characterized by monsoon in summer and westerlies[1] in winter. Summer months are wet, humid and hot while winter months are cold and mostly dry (Khadka et al., 2023; Sigdel et al., 2022). The study area receives highest rainfall between June to September.

Phewa Lake: Phewa Lake, the second-largest lake in Nepal, is located in the Pokhara Valley of Kaski District. It lies at an altitude of 742 m above sea level and covers an area of approximately 4.43 km² (Dahal, 2018). The areas surrounding Phewa Lake, is home to diverse ethnic groups, including the Gurung, Magar, Brahmin, Chhetri, and Newar communities (Gautam, 2019). The lake supports various livelihoods, particularly through agriculture, tourism, fisheries, and local handicrafts. A study by Paudel (2021) found that 65% of households in the vicinity of Phewa Lake are directly or indirectly dependent on tourism-related business in the area. The introduction of non-native plants into the Phewa wetland, the removal of buffering vegetation, are the major threats to lake upland forest deterioration and unplanned urbanization (Dixitet al., 2017; Pathak et al., 2021a).

Begnas Lake: Begnas Lake, the third-largest lake in Nepal, is situated in the southeastern part of the Pokhara Valley in Kaski District. It lies at an altitude of 650 m above sea level and covers an area of approximately 3 km² with maximum depth of 12.5 m (Thakuri et al., 2021) Begnas Lake is a natural freshwater lake fed by rainwater and streams from the surrounding hills (Gurung, 2021). The Begnas Lake region is home to various ethnic communities, including the Gurung, Magar, and Dalit groups (Khadka, 2019). The lake supports diverse economic activities, primarily tourism, fishing, and agriculture. A study by Bhandari (2022) revealed that around 55% of households in the vicinity depend on lake-related occupations for their livelihoods.

Rupa Lake: Situated at an altitude of approximately 600 m above sea level, Rupa Lake covers an area of 1.35 km² (Gurung, 2021). Rupa Lake is a natural freshwater lake fed by streams originating from the surrounding hills and rainwater. The Rupa Lake region is home to diverse ethnic communities, including Gurung, Magar and Dalit groups (Khadka, 2020). The lake supports various economic activities, particularly fisheries, agriculture, and ecotourism. A study by Bhandari (2022) indicated that approximately 50% of households in the vicinity depend on lake-related occupations for their livelihoods. The wetland of Rupa Lake is facing a lot of threats including invasive alien plant species,

[1]The westerlies refer to mid-latitude winds that blow from the west to east, typically between 30° and 60° latitude. In South Asia, they play a crucial role during winter and spring.

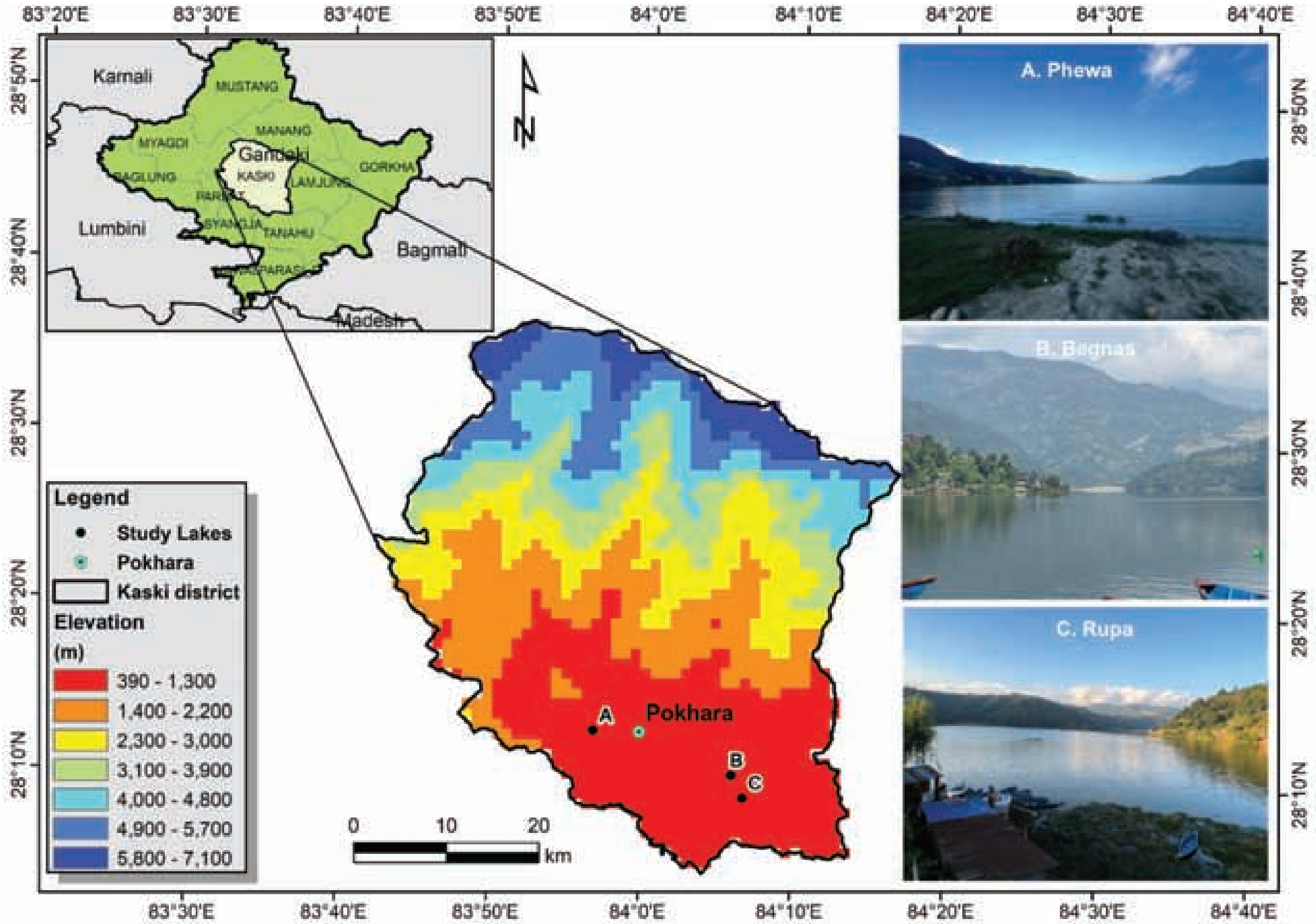

Figure 1: Overview of the study area and three lakes, **A.** Phewa Lake, **B.** Begnas Lake, **C.** Rupa Lake. The photos of the three lakes in side panel capture the visual images which are the key focus of this research

eutrophication, siltation, encroachment, settlement area expansion habitat destruction, depletion of species abundance and diversity, loss of economic integrity (Gautam et al., 2019; Pokhrel et al., 2019).

Field visit, data collection and analysis

The study sites were visited twice in 2023, covering two distinct seasons corresponding to the peak flowering periods of wetland flora: the monsoon season (June-August) and the autumn season (September-November). During these field visits, specimens and photographs of key plant species were collected. Voucher specimens of flowering plants were gathered from along the lakeshores, within the lakes, and surrounding areas. Free-floating and submerged species located within the lakeshore zones were collected following the methodology of Haynes (1984).

The collected specimens were pressed and dried using newspapers and herbarium presses. Identification was carried out with reference to regional and national floras, including Grierson & Long (1983-2001), Wu et al. (1994-2008), and Watson et al. (2011). Additionally, the samples were cross-verified with collections housed at the National Herbarium and Plant Laboratories (KATH) and the Tribhuvan University Central Herbarium (TUCH). Expert opinions were sought for species with uncertain identification. Plant family names were assigned according to the APG (Angiosperm Phylogeny Group)-IV system (Chase et al., 2016), while Roskov et al. (2020) was used for genus and species nomenclature.

After the identification of plant species, their use, status and availability in three lakes were discussed through KII and FGD. Status of plants availability was also derived from people's perception.

Key Informants' Interview (KII): KII was used to target representatives from among the community leaders, representatives from the IPLC groups, women's networks and local government representatives. A total of 91 individuals were selected for conducting KII. A snowball sampling technique was used to purposively select individuals who are assumed to have traditional knowledge on the utilization of plant resources. Aged and experienced person having more than 30 years' of experience (especially local healers or elderly people) were selected as key informants. A checklist and semi-structured questionnaire were employed as tools to conduct Key Informant Interviews (KIIs), facilitating the collection of relevant quantitative data.

Focus Group Discussion (FGD): Three Focused Group Discussions (FGDs) were organized with Lake Conservation and Management Committees of Phewa, Rupa, Begnas lake basins. A total of 30 representatives from major stakeholder groups - Lake Management Committees, Community Forest User Groups, Lake Conservation Committee, Boater's Association, Fishing Cooperatives, Mother Groups, Local Youth Club and others participated in the FGDs. Mostly elder persons living at the nearby settlements of the lake clusters especially belonging to Water User Group, Informal Irrigation User Group, Fishermen Group, Lake Dependent Communities (Jalahari) participated in the focused group discussion. Checklists were used as a dialogue tool for conducting FGD ensuring that all the issues related to the utilization of the floral biodiversity in the lake basins.

Results and Discussion

Floral diversity and use categories

A total of 81wetland-plant species belonging to 72 genera, and 42 families generally used by indigenous communities were recorded in the lake basins of Phewa, Begnas, and Rupa. Asteraceae family recorded highest number (9) followed by Fabaceae (6) and Poaceae (5) in study area (Figure 2, Appendix 1).

The local communities of the study areas have a long tradition of using the plant resources of wetlands

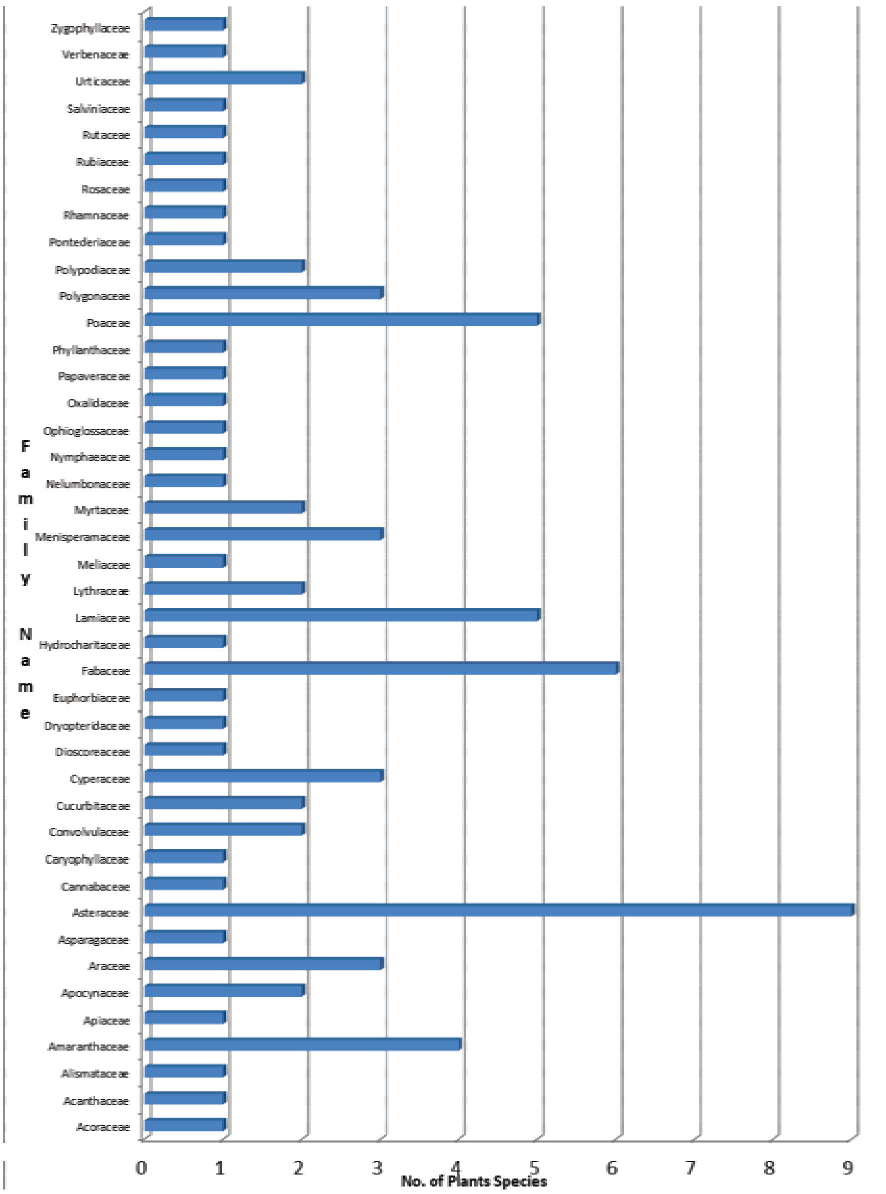

Figure 2: Family categorization of plants species in Phewa, Begnas and Rupa lakes

to meet their various basic needs such as food, medicine, fodder, firewood and other miscellaneous uses. Out of the total 81 species documented, 39 plant species are used for medicinal purposes, while 6 plants species are used for food and 4 plants species used for fodder purposes in all three lakes basin area. Likewise,10 plants species are used for both medicine and food purposes and 5 plants species used for medicine and fodder while only 2 plants species used for firewood purpose in all three-lake basin area of Phewa, Begnas and Rupa lake (Table1).

Growth form of the plants

Recorded plant species from the study area also categorized according to their growth form. It was found that 88.9% percent of plants species are in emergent form, 7.4% are in free floating form, 2.5% are in floating leaved rooted form and only one plant species was available in submerged condition (Figure 3).

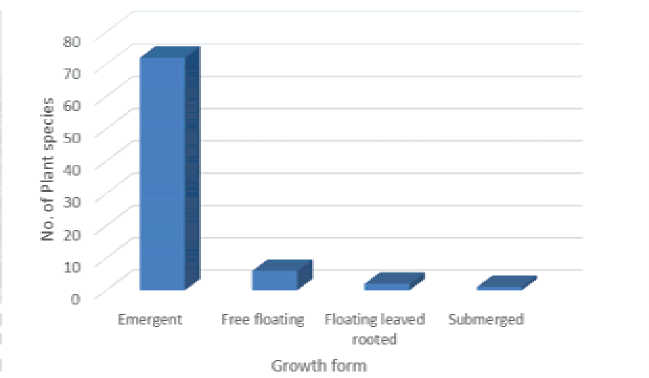

Figure 3: The growth form of the species

The status of the recorded plant species was discussed in KII and FGD with local people. Based on their perceptions, 18 plant species were reported to be in decline, while 6 species were noted to be increasing. The remaining 57 species were considered stable, out of a total of 81 plant species documented in the study lakes (Figure 4, Appendix 1).

South Asian wetlands are facing severe stress from human activities that are resulting habitat degradation and biodiversity decline (Trisal, 2023). These unsustainable anthropogenic activities are disrupting impacting structure and functions especially impacting aquatic and surrounding species leading to overall decline in wetland ecosystem health characterized by ecosystem conversion and degradation. Change in land and water use, water discharge, ecosystem structure and function can all have profound effects on the functioning of a lake (Olubode et al., 2011). Rapid invasion by exotic species are threatening the native species and affecting the status of a lake flora (Palmik et al., 2013).

Most of these invasive species have appeared in the last 20 years of period and are heavily increasing in recent years reinforcing the role of anthropogenic factors exacerbated by climate change. Pathak et al. (2021b) also described the invasiveness as major threats for declining the native species.

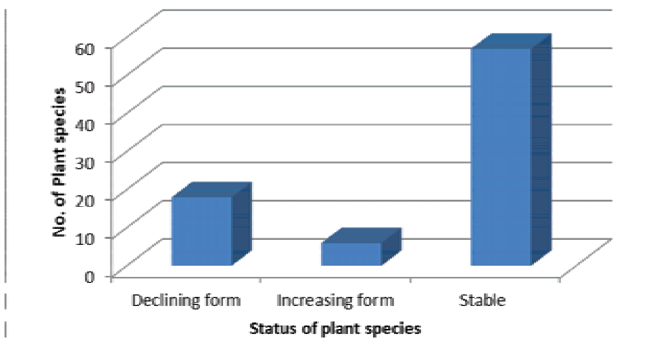

Figure 4: Status of plant species in three lake basin area of Phewa, Begnas and Rupa lake

Table 1: Plants used category in Phewa, Begnas and Rupa lakes

S.N.	Uses	Total used	Phewa Lake	Begnas Lake	Rupa Lake
1.	Medicine	39	34	30	38
2.	Food	6	5	6	6
3.	Fodder	4	3	3	4
4.	Firewood	2	2	2	2
5.	Medicine and Food	10	9	8	9
6.	Medicine and Fodder	5	5	4	4

Ageratina adenophora (Spreng.) R.M.King&H. Rob., *Chromolaena odorata* (L.) R.M.King&H. Rob., *Lantana camara* L., *Mikania micrantha* Kunth, *Pontederia crassipes* Mart. and *Ipomoea carnea* subsp. *fistulosa* (Mart. ex Choisy) D.F.Austin are considered as alien species with high threat to native species and ecosystems. Similarly, species like among these the *Pontederia crassipes* Mart., *Ipomoea carnea* subsp. *fistulosa* (Mart. ex Choisy) D.F.Austin, *Mikania micrantha* Kunth, *Alternanthera philoxeroides*(Mart.) Griseb., *Pistia stratiotes* L. are major IAPS seriously invaded in wetlands of Nepal (Ministry of Forest and Soil Conservation/Conservation and Sustainable Use of Wetlands Nepal [MoFSC/CSUWN], 2011). Lakes in Pokhara are already severely invaded by water hyacinth (Gautam et al., 2019). This study found that invasive alien species like *Pontederia crassipes* Mart. are impacting all the three lakes although their impact is relatively high in Phewa Lake as compared to other lakes (Pathak et al., 2021a). It was also found that rapid rate of human encroachment, increased eutrophication, invasive species, water diversion, toxic contamination, acidification, unregulated fishing, sedimentation, sewage water pollution from rapidly mushrooming hotels and the effects of extreme weather occurrences are some of the major threats to Phewa Lake (Rai, 2000).

Invasive species are actively invading, creating a danger to increasing agricultural operations, forest, culture, transportation, trade, power production, recreation, and fisheries by eradicating and substituting native biodiversity. Due to sedimentation caused by soil erosion, landslides, and significant water hyacinth invasion, the lake and its watershed are found to be under intense and exhausting pressure for the last several decades confirming the finding of similar study (Rai, 2000). Likewise, this study also aligns with the finding by Lama et al. (2018) who recorded impact of invasive alien species impact in Begnas lake. The loss of aquatic biodiversity has been exacerbated by the serious challenges posed by the invasion of foreign species such as water hyacinth which is especially difficult to control. Similarly, Pathak et al. (2021a) also describe terrestrial invasive plants such as *A. adenophora, P. stratiotes,* and *I. carnea* species are posing similar challenges.

Conclusion

The study has found a rich diversity of plant species used by Indigenous Peoples and Local Communities (IPLCs) in the three major lake basins of Pokhara valley. The study particularly highlights the utilization status of wetland basin flora of the study area. The IPLCs living around the three major lakes of Pokhara Valley have a long history of high dependency on the lake basin's rich floral diversity to meet their diverse livelihood needs, cultural preservation pursuits, and traditional medicinal practices. These lakes and their surrounding basin are found to harbor a variety of plant species used for food, fodder, fuelwood, and herbal medicines. Local communities, including the ethnic communities especially, Gurung, Magar and Jalarib peoples have developed extensive knowledge on utilization of plants.

However, spreading invasive alien species are threatening the sustainability of these plant resources ultimately having impacts on Indigenous Peoples and Local Communities (IPLC)'s livelihood and culture. Efforts to document and preserve traditional botanical knowledge, alongside locally-led conservation initiatives, are crucial for maintaining the ecological balance between wetlands and dependent communities thereby sustaining local livelihoods and bio-cultural heritage. Integrating indigenous knowledge with modern knowledge and community-led conservation strategies can help protect these valuable aquatic and terrestrial plant resources while promoting sustainable use of wetland biodiversity and ecosystem resources.

Author Contributions

K P Sigdel contributed in data collection, analysis and manuscript preparation and the co-authors- N P Ghimire and M B Karki significantly contributed in conceptualization, editing, framework development and analysis. P Shrestha contributed in data collection and tabulation.

Acknowledgements

Authors would like to thank to local communities of Phewa, Begnas and Rupa Lakes for providing information on traditional usage of plant resources. We'd like to thank to Nepal Academy of Science and Technology (NAST), Ministry of Forests and Environment (EBA-II Project) for providing partial financial support.

References

Berkes, F., Colding, J., & Folke, C. (2000). Rediscovery of traditional ecological knowledge as adaptive management. *Ecological Applications, 10(5),* 1251-1262.

Bhandari, K. (2022). Economic impact of ecotourism on local communities: A case study of Rupa Lake. *Nepal Journal of Economics*, *15*(2), 67-85.

Brokensha, D., Warren, D., & Werner, O. (1980). *Indigenous knowledge systems and development.* University Press of America.

Brosi, B. J., Balick, M. J., Wolkow, R., Lee, R., Kostka, M., Raynor, W., Gallen, R., Raynor, A., Raynor, P., & Ling, D. L. (2007). Cultural erosion and biodiversity: Canoe-making knowledge in Pohnpei, Micronesia. *Conservation Biology, 21* (3), 875-879.

Central Bureau of Statistics. (2014). *National population and household survey, 2011*. National Planning Commission, Government of Nepal.

Chase, M. W., Christenhusz, M. J., FAY, M. F.; Byng, J. W., Judd, W, S., Soltis, D. E., & Stevens, P. F. (2016). An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG IV. *Botanical Journal of the Linnean Society, 181*(1), 1-20.

Dahal, M. (2018). *Hydrological and ecological study of Phewa Lake*. Himalayan Research Institute.

Department of Hydrology and Meteorology. (2017). Observed climate trend analysis in the of Nepal (1971-2014).

Dixit, A., Karki, M., & Shukla, A. (2015). *Vulnerability and Impacts Assessment for Adaptation Planning in Panchase Mountain Ecological Region, Nepal*. Government of Nepal, United Nations Environment Programme, United Nations Development Programme, International Union for Conservation of Nature, German Federal Ministry for the Environment, Nature Conservation, Building and Nuclear Safety and Institute for Social and Environmental Transition-Nepal. https://www.undp.org/sites/g/files/zskgke326/files/migration/np/EBA-vulnerability_impacts_assessment.pdf

Donlan, C. J., Tershy, B. R., Campbell, K., & Cruz, F. (2003). Research for requiems: The need formore collaborative actions in eradication of invasive species. *Conservation Biology*, *17*(6), 1850-1851.

Egeru, A. (2012). Role of indigenous knowledge in climate change adaptation: A case study of Teso sub-region eastern Uganda. *Indian Journal of Traditional Knowledge*, *11* (2), 217-224.

Gautam, D., Bhattarai, S., Sigdel, R., Jandng, C. M B., Mujahid, A., & G. C, D. B. (2019). Climate variability and wetland resource in Rupa Lake Catchment, Nepal. *North American Academic Research*, *2* (11), 82-91. https://doi.org/10.5281/zenodo.3568477.

Gautam, R. (2019). *Demographic changes in Pokhara Valley: A sociological perspective*. Pokhara University Press.

Grierson, A. J. C., & Long, D. G. (1983-2001). *Flora of Bhutan*. Royal Botanic Garden, Edinburgh.

Gurung, M. (2020). *Hydrology and ecology of Begnas Lake*. Himalayan Research Institute.

Guragain, G. P. (2024). Progress of diversity and inclusion in Nepal. *Damak Campus Journal, 13* (1), 1-14

Gurung, M. (2021). *Hydrology and ecology of Rupa Lake*. Himalayan Research Institute.

Haynes, R. R. (1984). Techniques for collecting aquatic and marsh plants. *Annals of the Missouri Botanical Garden*, *71*(1), 229-231.

International Centre for Integrated Mountain Development (2007). *Local knowledge for disaster preparedness*.

Karki, M. B. (2017). *Challenges, opportunities and trade-offs in commercialization of medicinal and aromatic plants in South Asia Region*.

Karki, M. (2015). Challenges, opportunities and trade-offs in commercialization of medicinal and aromatic plants in South Asia region. *In Invited paper presented at the workshop on current challenges and recommendations.* Government of India. https://www.Academia.Edu/12863952/Challenges_Opportunities_And_Trade-Offs_In_Commercialization_Of_Medicinal_And_Aromatic_Plants_In_South_Asia_Region

Karki, M., Pokhrel, P., & Adhikari, J. R. (2017). Climate change: Integrating indigenous and local knowledge into adaptation policies and practices: A case study from Nepal. *Shifting Cultivation Policies: Balancing Environmental and Social Sustainability*. CABI.

Karki, M., & Adhikari, J. R. (2015). Integrating indigenous, local and modern knowledge for sustainable conservation and management of forest ecosystems in Nepal. *Forestry Nepal: Gateway to Forestry Information in Nepal*. https://www.forestrynepal.org/publications/article/6307

Khadka, B. (2019). *Ethnic diversity and socioeconomic development in Pokhara Valley*. Pokhara University Press.

Khadka, B. (2020). *Ethnic diversity and socioeconomic development in Pokhara Valley*. Pokhara University Press.

Khadka, N., Chen, X., Sharma, S., & Shrestha, B. (2023). Climate change and its impacts on glaciers and glacial lakes in Nepal Himalayas. *Regional Environment Change*, *23*, 143. https://doi.org/10.1007/s10113-023-02142-y

K. C., J., Gurung, K., & Shrestha, P. (2014). Lowland wetlands in Nepal. *The Initiation*, *5*. 10.3126/init.v5i0.10269.

Lama, F., Thakuri, S., Ghimire, N. P., & Malla, R. (2018). Ecotone vegetation and water quality of Rara and Begnas Lakes, Nepal. *Himalayan Biodiversity*, *6*, 27-37. 10.3126/hebids.v6i0.33529.

Loh, J., & Harmon, D.(2014). *Biocultural Diversity: Threatened Species, Endangered Languages*. WWF Netherlands .

Mercon, J., Vetter, S., Tengo, M., Cocks, M., Balvanera, P., Rosell, J.A., Ayala-Orozco, & B. (2019). From local landscapes to international policy: Contributions of the biocultural paradigm to global sustainability. *Glob. Sustain. 2*, e7.

Ministry of Forest and Soil Conservation/ Conservation and Sustainable Use of Wetlands Nepal. (2011). *Wetlands invasive alien species management guidelines*. Conservation and Sustainable Use of Wetlands Nepal Project, Ministry of Forests and Soil conservation, Nepal.

Ministry of Forests and Soil Conservation (2006). *Third national report to the Convention on Biological Diversity.*

Ministry of Education, Science and Technology (2015). *Indigenous and local knowledge and practices for climate resilience in Nepal, mainstreaming climate change risk management in development.*

National Lake Conservation Development Committee (2021). *Inventory of lakes in Nepal (Main Report*) (pp.145). National Lake Conservation Development Committee (NLCDC): Ministry of Forests and Environment: Government of Nepal.

Olubode. O. S., Awaodoyin, R. O., & Ogunyemi, A. (2011). Floral diversity in the wetlands of Apete River, Eleyele lake and Oba Dam in Ibadan, Nigeria: Its implication for biodiversity erosion. *West African Journal of Applied Ecology, 18*, 21-35.

Palmik, K., Maemets, H., Haldina, M., & Kangur, K. (2013). A comparative study of macrophyte species richness in differently managed shore stretches of lake Peipsi. *Limnologica, 43*, 245-253.

Pathak H. N., Bhuju, D. R., Shrestha B. B., & Ranjitkar, S. (2021a). Impacts of invasive alien plants on ecosystem services of Ramsar lake cluster in middle mountain Nepal. *Global Ecology and Conservation*, *27*, e01597.

Pathak, H. N., Shrestha, B. B., Bhuju, D., & Subedi, S., (2021b). Spatial distribution of invasive alien plants in Pokhara valley, Nepal. *Pakistan Journal of Botany*, *5*3. 10.30848/PJB2021-3(2).

Paudel, S., (2021). The socioeconomic impact of tourism in Pokhara: A case study of Phewa Lake. *Journal of Nepalese Studies*, *10*(1), 45-60.

Pokhrel, H. R., Gautam, D., Jandug C. M., Bhattarai, S., Baral, K., Bhandari, J., & Gahatraj, G. (2019). Status and diversity of invasive alien plant species in Rupa Lake, Kaski District, Nepal. *International Journal of Emerging Technologies and Innovative Research*, *6*(6),1028-1035.

Rai, A. K. (2000). Limnological characteristics of subtropical Lakes Phewa, Begnas, and Rupa in Pokhara Valley, Nepal. *Limnology*, *1*(1) 33-46. https://doi.org/10.1007/s102010070027.

Reaser, J. K., Meyerson, L. A., Cronk, Q., De Poorter, M. A. J., Eldrege, L. G., & Green, E. (2007). Ecological and socio-economic impacts of invasive alien species in island ecosystems. *Environmental Conservation*, *34*(2), 98- 111.

Roskov, Y, Ower, G., Orrell, T., Nicolson, D., Bailly, N., Kirk, P. M., Bourgoin, T., Dewalt, R. E., Decock, W., Nieukerken, E., & Penev, L.E. (Eds.). (2020). *Species 2000 & ITIS Catalogue of Life, 2020.*

Sigdel, K. P., Ghimire, N. P., Pandeya, B., & Dawadi, B. (2022). Historical and projected variations of precipitation and temperature and their extremes in relation to climatic indices over the Gandaki River Basin, Central Himalaya. *Atmosphere*, *13*, 1866 https://doi.org/10.3390/atmos13111866.

Siwakoti, M. (2012). Threats and opportunity of invasive alien plant species in wetlands conservation of Nepal. *Proceedings of International Wetland Symposium*, 66-72.

Thakuri, S., Lama, F., Malla, R., Khadka, N., Ghimire, N. P., & Salerno, F. (2021). Lake watershed dynamics and bathymetry modeling of Rara and Begnas Lakes in Nepal. *Earth*, *2* (2), 272-286. https://doi.org/10.3390/earth2020016

Trisal, C. (2023). Wetlands of Hindu Kush Himalayas - ecosystem functions, services and implications of climate change. https://www.icimod.org/resources/77

Watson, M. F., Mkiyama, S., Pendry,C., Rajbhandari, K. R., & Shrestha, K. K. (2011). *Flora of Nepal* (Vol.3). The Royal Botanic Garden Edinburgh.

Wu, Z., Raven, P. H., & Hong, D. (Eds.). (1994-2008). *Flora of China* (Vol. 4, 8, 11, 13, 14, 16, 18, 19, 20, 22, 24, 25). Science Press and Missouri Botanical Garden Press.

Appendix 1: Status of plants in Phewa, Begnas and Rupa Lake basin area having ethnobotanical uses

S.N.	Scientific name	Family	Local Name	Availability			Used value	Growth form	Plant Status	Specimen No.
				Phewa	Begnas	Rupa				
1	*Acorus calamus* L.	Acoraceae	Bojho	√	√	√	Me	E	D	PL 10
2	*Ageratina adenophora* (Spreng.) R.M.King & H.Rob.	Asteraceae	Banmara	√	√	√		E	I	PL 56
3	*Ageratum conyzoides* L.	Asteraceae	Ganmane Ghans	√	√	√		E	I	BL 11
4	*Ageratum houstonianum* Mill.	Asteraceae	Nilo Gandhe	√		√		E	S	PL 23
5	*Alstoniascholaris* (L.) R.Br.	Apocynaceae	Chatiwan	√	√		Me	E	D	PL 27
6	*Alternanthera philoxeroides* (Mart.) Griseb.	Amaranthaceae		√	√	√		E	I	PL 16
7	*Alternanthera sessilis* (L.) DC.	Amaranthaceae	Bhringijhar			√	Me	E	S	BL 40
8	*Amaranthus spinosus* L.	Amaranthaceae	Kande Lude	√	√	√	Me	E	S	BL 30
9	*Argemone mexicana* L.	Papaveraceae	Kandejhar	√	√	√		E	S	BL 55
10	*Artemisia indica* Willd.	Asteraceae	Titepati	√	√	√	Me	E	S	PL 09
11	*Arundo donax* L.	Poaceae	Narkat	√	√	√	Fdr	E	S	BL 32
12	*Asparagus racemosus*Willd.	Asparagaceae	Kurilo	√		√	Me, Fd	E	D	BL 48
13	*Azolla imbricata* (Roxb.) Nakai	Salviniaceae	Pani uneu	√	√	√	Me	FF	D	PL 18
14	*Berchemia edgeworthii* M.A. Lawson	Rhamnaceae	Angeri	√	√	√	Fd	E	S	BL 60
15	*Botrychium lanuginosum* Wall. ex Hook & Grev.	Ophioglossaceae	Jaluko	√	√	√	Me	E	S	RL 55
16	*Callicarpa macrophylla* Vahl	Lamiaceae	Daikamli	√	√	√	Me, Fd	E	S	RL 46
17	*Cannabis sativa* L.	Cannabaceae	Gaanja, Bhang	√		√	Me	E	S	PL 31
18	*Castanopsis indica* (Roxb. ex Lindl.) A.DC.	Fagaceae	Katus	√	√	√	Fd	E	S	RL 60
19	*Centella asiatica* (L.) Urb.	Apiaceae	Ghodtapre	√	√	√	Me	E	D	PL 51
20	*Chenopodium album* L.	Amaranthaceae	Bethe; Batuwa	√	√	√	Me, Fd	E	S	PL 13
21	*Chromolaena odorata* (L.) R.M.King & H.Rob.	Asteraceae	Banmara	√	√	√	Fr	E	S	PL 14
22	*Citrus medica* L.	Rutaceae	Bimiro		√	√	Fd	E	D	PL 15
23	*Cissampelos pareira* L.	Menispermiaceae	Batulpate	√		√	Me	E	S	RL 78
24	*Colebrookea oppositifolia* Sm.	Lamiaceae	Dhurseli	√	√	√	Me	E	S	RL 37
25	*Cyperus esculentus* L.	Cyperaceae	Mothe	√	√	√		E	S	PL 19
26	*Cyperus rotundus* L.	Cyperaceae	Mothe	√	√	√	Fdr	E	S	PL 11
27	*Desmostachyabipinnata* (L.) Stapf	Poaceae	Kush	√	√	√	Me, Fdr	E	S	PL 74

S.N.	Scientific name	Family	Local Name	Availability			Used value	Growth form	Plant Status	Specimen No.
				Phewa	Begnas	Rupa				
28	*Dioscorea bulbifera* L.	Dioscoreaceae	Bhyakur	√	√	√	Me	E	S	PL 71
29	*Drymaria cordata* (L.) Willd. ex Schult.	Caryophyllaceae	Abhijalo	√		√	Me	E	D	RL 72
30	*Dryopteris* spp	Polypodiaceae	Pani Neuro	√	√	√	Fd	E	S	PL 45
31	*Dryopteris cochleata* (D.Don) C.Chr.	Polypodiaceae	Kalo Neuro	√	√	√	Me, Fd	E	S	RL 73
32	*Ecliptaprostrata* (L.) L.	Asteraceae	Bhringiraj	√	√	√	Me	E	S	PL 78
33	*Gerardianadiversifolia*	Urticaceae	Chalnesisnu	√	√	√	Me	E	S	BL 20
34	*Hydrilla verticillata* (L.f.) Royle	Hydrocharitaceae	Panijhyau, Sewar,	√		√	Me	S	D	BL 65
35	*Ipomoea aquatica* Forssk.	Convolvulaceae	Kalami sag	√	√	√		FF	S	BL 26
36	*Ipomoea carnea* subsp. *fistulosa* (Mart. ex Choisy) D.F.Austin	Convolvulaceae	Besaram	√	√	√		FF	S	PL 18
37	*Justicia adhatoda* L.	Acanthaceae	Asuro	√	√	√	Me	E	S	RL 58
38	*Leersiahexandra* Sw.	Poaceae	*KarauteJhar*	√	√	√	Me	E	S	RL 57
39	*Lemna minor* L.	Araceae	Leu	√	√	√	Me	FF	S	PL 42
40	*Lantana camara* L.	Verbenaceae		√	√	√	Fr	E	I	BL 25
41	*Melia azedarach* L.	Meliaceae	Bakaino	√	√	√	Me, Fdr	E	S	RL 79
42	*Mikania micrantha* Kunth	Asteraceae	Aalupate	√	√	√	Fr	E	I	BL 28
43	*Millettia extensa* (Benth.) Benth. ex Baker	Fabaceae	Gaujo		√	√	Me	E	S	PL 81
44	*Mimosa pudica* L.	Fabaceae	Lajjabati	√	√	√	Me	E	S	BL 72
45	*Nelumbo nucifera* Gaertn.	Nelumbonaceae	Seto Kamal	√		√	Me	FF	D	PL 08
46	*Nephrolepis cordifolia* (L.) C.Presl	Polypodiaceae	Pani amala	√	√	√	Me, Fd	E	S	BL 73
47	*Nymphaea rubra Roxb. ex Salisb.*	Nymphaeaceae		√		√	Fdr	E	S	PL 78
48	*Oryza rufipogon* Griff.	Poaceae	Nabo Dhan			√	Me	E	D	BL 31
49	*Oxalis corniculata* L.	Oxalidaceae	Chariamilo	√	√	√	Me	E	S	RL 71
50	*Parthenium hysterophorus* L.	Asteraceae	Pati	√	√	√		E	S	PL 46
51	*Periploca calophylla* (Wight) Falc.	Apocynaceae	Sikari Lahara	√	√	√	Me	E	D	RL 44
52	*Phyllanthus emblica* L.	Phyllanthaceae	Amala	√	√	√	Me, Fd	E	D	RL 48
53	*Persicariabarbata* (L.) H.Hara	Polygonaceae	Pirre jhar	√	√	√	Me	E	S	BL 30
54	*Persicaria hydropiper* (L.) Delarbre	Polygonaceae	Pirre jhar	√	√	√	Me	E	S	RL 49
55	*Pistia stratiotes* L.	Araceae	Banda ghass	√	√	√		E	S	PL 62
56	*Pogostemon benghalensis (Brum.f.) Kuntze*	Lamiaceae	Gudheli	√	√	√	Me	E	S	BL 46

S.N.	Scientific name	Family	Local Name	Availability			Used value	Growth form	Plant Status	Specimen No.
				Phewa	Begnas	Rupa				
57	*Pontederia crassipes* Mart.	Pontederiaceae	Jalakumbhi	√	√	√		FF	I	PL 49
58	*Premna barbata* Wall. ex Schauer	Lamiaceae	Gidari	√	√	√	Me, Fdr	E	S	BL 68
59	*Rhaphidophora glauca* (Wall.) Schott	Araceae	Haddijor	√		√	Me	E	D	PL 06
60	*Rubus ellipticus* Sm.	Rosaceae	Ainselu	√	√	√	Me, Fd	E	S	BL 18
61	*Rumex nepalensis* Spreng.	Polygonaceae	Halhale		√	√	Me	E	S	BL 37
62	*Sagittariaguayanensis* subsp. *guayanensis*	Alismataceae	Karkalejhar	√	√	√	Me	E	S	PL 20
63	*Falconeria insignis* Royle	Euphorbiaceae	Khirro	√	√		Me	E	S	BL 35
64	*Schoenoplectiellamucronata* (L.) J.Jung & H.K.Choi	Cyperaceae	Gud mothe	√	√	√		E	S	RL 45
65	*Senna occidentalis* (L.) Link	Fabaceae	Taprejhar	√	√	√		E	S	PL 67
66	*Senna tora* (L.) Roxb.	Fabaceae	Taprejhar	√	√	√		E	S	BL 05
67	*Solena amplexicaulis* (Lam.) Gandhi	Cucurbitaceae	Golkakri	√			Me, Fd	E	S	RL 38
68	*Solena heterophylla* Lour.	Cucurbitaceae	Golkarki		√	√	Me, Fd	E	S	BL 59
69	*Spermacoce alata* Aubl.	Rubiaceae		√	√	√	Me	E	S	BL 39
70	*Stephania glandulifera* Miers	Menispermaceae	Gujargano	√	√	√	Me	E	D	BL 57
71	*Syzygiumcumini* (L.) Skeels	Myrtaceae	Jamun	√			Me, Fdr	E	S	PL 07
72	*Syzygium nervosum* DC.	Myrtaceae	Kyamun	√	√	√	Me, Fdr	E	D	PL 67
73	*Tinospora cordifolia* (Willd.) Hook.f. & Thomson	Menispermaceae	Gurjo	√	√	√	Me	E	D	BL 49
74	*Trapa natans* var. *bispinosa* (Roxb.) Makino	Lythraceae	Simalkade	√	√	√	Fd	FLR	D	BL 03
75	*Trapa natans* var. *quadrispinosa* (Roxb.) Makino	Lythraceae	Bhaisikade	√	√	√	Fd	FLR	D	PL 77
76	*Tribulus terrestris* L.	Zygophyllaceae	Gaikhure	√	√	√	Me	E	S	BL 02
77	*Trifolium repens* L.	Fabaceae	Beuli, Pyauli,	√	√	√	Me	E	S	BL 34
78	*Urtica dioica* L.	Urticaceae	Sisnoo	√	√	√	Me, Fd	E	S	BL 50
79	*Vetiveriazizznioides*	Poaceae	Usir		√	√	Fdr	E	S	PL 05
80	*Vitex negundo* L.	Lamiaceae	Simali Ghas		√	√	Me	E	S	BL 74
81	*Xanthium strumarium* L.	Asteraceae	Bhaise Kada	√	√	√	Me	E	S	BL 33

Note: Growth form (E= Emergent; FF= Free floating; FLR= Floating leaved rooted aquatics; Sub = submerged), Used Value (Me- Medicine, Fd- Food, Fdr- Fodder, Fr- Fire wood, I= Increasing, D= Decreasing, S= Stable)

Journal of Plant Resources (2025) 23(1), 80-90
https://doi.org/10.3126/bdpr.v23i1.83356

Ethnomedicinal Plants Used by the Dura Tribe in Lamjung District, Nepal

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Abstract

An ethnobotanical survey was conducted to document medicinal plants used by the Dura tribe for the treatment of various ailments in Neta, Madhyanepal municipality, Lamjung District, Nepal. Field visits were carried out from October 2022 to April 2023 to collect ethnobotanical data. Data were gathered from 56 key informants using semi-structured interviews, un-structural interviews, group discussions, and questionnaires. The study area is rich in ethnic diversity, encompassing a range of ethnic groups such as Gurung, Dura, Brahmin, Chhetri, Magar, Damai, Kami, Sarki, etc. A total of 73 plant species belonging to 45 families, used to treat 64 different ailments, were recorded. The factor of informant consensus (FIC) for muscular/skeletal disease category had the highest value. The results showed that four plant species namely: *Calotropis gigantea, Drynaria quercifolia, Hoya lanceolata* and *Periploca graeca* were culturally important to the Duras for the treatment of muscular sprain and bone fracture. Herbs were the most commonly used plant for medicine followed by tree and shrub. Leaves were the most frequently used plant parts, and the majority of plants were used in the form of juice. Wild plant species accounted for 63% of the medicinal plants used. Although the Duras are a marginalized tribe, they possess huge indigenous knowledge about medicinal plants. Moreover, phytochemical analysis of culturally valued plants within the Dura community should be conducted to validate this knowledge scientifically.

Keywords: Duradanda, Indigenous knowledge, Madhyanepal municipality, Muscular sprain, *Periploca graeca*

Introduction

Nepal is a multiethnic country with 142 caste/ethnicity (National Population & Housing Census [NPHC], 2021). These ethnic groups possess indigenous knowledge of medicinal plants, and numbers of plants are used as medicine exclusively by certain ethnic groups. Ethnomedicine is a valuable source of information regarding useful medicinal plants of indigenous people. It benefits humanity in terms of traditional pharmacopoeias.

The Dura are one of the ethnic groups originally reside in Duradanda, Lamjung. Dura's have their own language, religion, and their own type of health care practices. Ethnic communities develop their indigenous knowledge on the use and management of plants including medicinal plants through long years of interaction with their surrounding (Gebeyehu et al., 2024). Ethnobotany serves as a tool to find out such knowledge of indigenous people.

The study of ethnobotany in Nepal began with the publication of Medicinal and Food Plants of East Nepal by Banerji (1955), and was continued by many other researchers. Besides ethno-medicinal study conducted on different geographical areas of the country, extensive research has also focused on specific ethnic groups such as Sherpas (Bhattarai, 1989; Sacherer, 1979), Gurungs (Coburn, 1984), Tharus (Dangol & Gurung, 1991; Ghimire & Bastakoti, 2009; Manandhar, 1985), Tamang (Luitel et al., 2014; Manandhar, 1991), Rautes (Manandhar, 1998), Chepangs (Rijal, 2011; Tamang et at., 2017), Limbus (Limbu & Rai, 2013), Magars (Acharya, 2012; Nemkul et al, 2018; Poudel & Gautam, 2008; Singh et al., 2018; Thapa, 2012) and others. Till the date, no ethnobotanical study has been conducted on the Dura tribe.

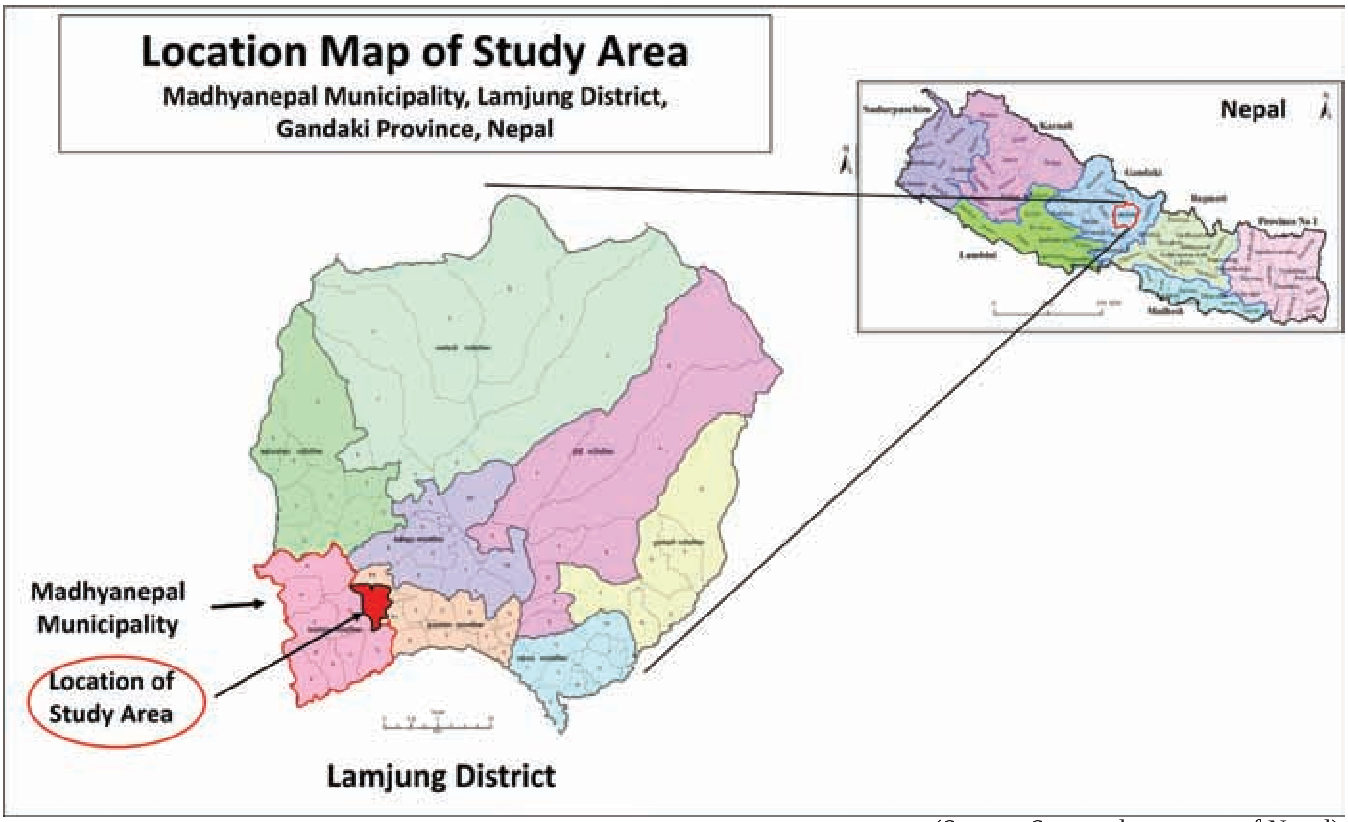
This current study is the first attempt to document the ethnomedicinal knowledge of the Dura tribe in Lamjung District. The total population of Duras in Nepal is 5581 (NPHC, 2021). It is very significant to study ethnomedicinal knowledge of the Duras before it is lost forever.

Materials and Methods

Study area

The study area lies in Lamjung District, Gandaki Province of Nepal (Figure 1). Lamjung District lies between 28º03'N to 28º30'N latitude and 84º11'E to 84º38'E longitude, and the Besisahar is it's headquarter. The elevation ranges from 385 m in the south to 8,162 m (Manaslu Himalaya) in the north above sea level (https://chinarinepal.com/ Lamjung). The temperature of the study area is hot in summer (June), with the average highest temperature around 35.7ºC, and cold and dry in winter (January), with the lowest average temperature around 14.5ºC and the least rainfall occurring in December. Madhyanepal municipality is one of the eight municipalities in Lamjung District. Various ethnic groups, including Gurung, Dura, Chhetri, Brahmin, Magar, Lama, Tamang, Kami, Damai, Sarki, reside in the study area.

The Dura tribe is an indigenous group from the Duradanda hill of Lamjung District. The population of Dura people throughout Nepal is 5,581 (NPHC, 2021), with2751 residing in Lamjung District. The Dura have their own language, known as the Dura language, and maintain their own religion and culture, though it is quite similar to that of the Gurung. There is history that at the end of fifteen century the Dura established a Shah ruler in Lamjung by removing the Ghale King (Adhikari, 1973-60). Local healers (Lamas) and elderly peoples hold the proper knowledge of medicinal plants used for healing practices. They have acquired wide range of traditional healing knowledge.



(Source: Survey department of Nepal)

Figure 1: Location map of the study area

Data collection

The study area was visited on October, 2022, January, 2023 and April, 2023. During the first visit in October 2022, researcher stayed for three days. This visit involved meeting with local people and community leaders to introduce the purpose of the research. It helped to recognize local traditional healers and people having knowledge on herbal formulations, for selection of key informants. The selection of informants must be purposive rather than random for the study of ethnobotany.

The next visit of study site was on January and April 2023, with one week stay in each visit. Necessary materials such as polythene bags, number tags, old news papers, plant press, hard boards, strings, plant cutter, digging tools, a camera, field notebook, markers, pencils were taken along during the field visits.

Ethnomedicinal information was collected through questionnaires, structured and un-structured interviews among healers and knowledgeable people (56 Key informants). Voucher Herbarium Specimens were prepared, identified, and confirmed by comparing them with authenticated specimens at National Herbarium and Plant Laboratories, Godawari (KATH). The prepared herbaria were deposited at Botany Department, Tri-Chandra Multiple Campus, Ghantaghar.

Factor for informant consensus (FIC)

The FIC was calculated as the number of use citations in each category (Nur) minus the number of species used (Nt), divided by the number of use citations in each category minus one (Heinrich et al., 1998):

$$\text{FIC} = \frac{\text{Nur} - \text{Nt}}{\text{Nur} - 1}$$

In order to use this tool, the illnesses were classified into broad disease categories (several diseases based on the organ systems in one category) such as: (1) gastrointestinal, (2) dermatological, (3) respiratory, (4) muscular/skeletal, (5) pain, (6) urinary tract infection (UTI), (7) blood related problems, (8) eye problems, (9) jaundice, (10) kidney stones, and (11) other.

Results and Discussion

Ethnomedicinal knowledge among the people in the study area

The present study recorded 73 plant species belonging to 47 families, which were used for the treatment of various ailments by the Dura tribe in the study area, with plants names provided in the Dura language (Appendix 1). Ethnomedicinal data were collected by interviewing 56 informants. Asteraceae was the most prominent family, with six species, followed by Lamiaceae (five species), Moraceae (four species), Apocynaceae, Fabaceae, Menispermaceae, and Rutaceae (each with three species), six families with two species and remaining 34 families with one species. Of the 73 plant species recorded, 37% (27 plant species) were cultivated and 63% (46 plant species) were wild.

Similarly, Manandhar (1987) reported Asteraceae as the most commonly used family for treatment of ailments among tribes of Lamjung District.

Leaves were the most commonly used plant part for medication. Leaf of 38 plant species of medicinal plants was used, followed by the root of 24 species, fruit of 21 species, bark of 17 species, stem of nine species, flower of eight species, whole plant of eight species, rhizome of five species, sap of four species, seed of four species, frond of one species and the latex of one species.. Various ailments were treated using different components of same plant, and multiple parts of the same plant were recorded to be used for treating various ailments.

The most commonly used form of preparation was juice (26%), followed by decoction (25%), paste (19%), powder (12%), chewing (10%), infusion (2%) and others.

In the Dura language *Bombax ceiba* L., *Psidium guajava* L., *Nephrolepis cordifolia* (L.) C. Presl *Phyllanthus emblica* L., and *Urtica dioica* L. are called as Eku, Basa, Naisu, Kon and Koke (Appendix 1) respectively. In contrast, in the Gurung language they are called as Chongonchhi, Belauti, Kyudabi, Titi and Pulu (Manandhar, 2002) respectively.

Juice mode of drugs were used for immediate effects and sometimes used as antiseptic (e.g.,eye drops), likely due to easy availability of fresh plants materials (Nemkul, 2022). In most cases, juice was the most preferred form of medication. Similar findings have been reported previously (Bastakoti, 2019; Bhandari et al., 2023; Bhattarai, 2017; Manandhar, 1987; Singh et al., 2012).

The use of latex of *Calotropis gigantea* (L.) Dryand to treat sprains, fractures and muscular pain is supported by previous study in Lamjung District (Manandhar, 1987). Tamang et al.(2017) reported similar medicinal activity practiced used by Chepang community.

The Duras of the study site used *Zanthoxylum armatum* DC. to treat toothache, dysentery and common cold, similar to previous reports (Acharya, 2012; Manandhar, 1987).

They also used *Justicia adhatoda* L. to treat tonsillitis, cough and asthma, aligning with findings from other studies (Bhandari et al., 2023; Tamang et al., 2017).

The Duras of the study site used *Woodfordia fruticosa* (L.) Kurz. for the treatment of dysentery and gastritis, similar to its previously reporting use in the Magar community (Nemkul et al., 2022)

Factor for informant consensus (FIC)

A total of 73 plant species were used for the treatment of 64 ailments, which were grouped into 11 different disease categories based on the human body parts affected by an illness, following Frei et al. (1998) with some modifications. The categories were: (1) gastrointestinal (diarrhea, helminthiasis, dysentery, gastritis, abdominal bloating, indigestion, intestinal ulcer, and dyspepsia), (2) respiratory (common cold, cough, tonsillitis, asthma, sinusitis, sore throat, and tuberculosis), (3) dermatological (burn, cuts, wounds, pimple, skin diseases, acne, scabies, furuncle, and vitiligo), (4) muscular/skeletal (sprain, and fracture, joint pain), (5) pain (body pain, headache, stomach pain, back pain, toothache, chest pain, rheumatoid arthritis, and gout.), (6) urinary tract infection (UTI) (urine infection, urine blockage) , (7) blood related problems (blood pressure problem, menstrual problem, Blood cancer and anemia), (8) eye problems (9) kidney stones (10) jaundice and (11) other (malaria, fever, nausea, paralysis, bee sting, snake bite, hair fall, posterior nose bleed, miscarriage/ problem in postpartum discharge, diabetes, epilepsy, piles, uric acid, mastitis, heart disease, liver disease, bone stuck in throat, and loss of appetite).

The number of taxa used for treatment of each disease category and the total number of use-reports were analyzed, and it was found that the factor for informant consensus (Fic) value (Table 1) ranged between 0.85 to 0.97. FIC for muscular/skeletal disease category had highest values (Fic 0.97), followed by dermatological troubles (Fic 0.96), gastrointestinal disorder and respiratory problems each (Fic 0.95), pain and other problems both (Fic 0.94), blood related problems (Fic 0.93), urinary tract infection (Fic 0.92), jaundice (Fic 0.92), kidney stone (Fic 0.87), and eye problems (Fic 0.85). The data showed high use reports of the medicinal plants by the Duras (Table 10).

Some other scientists (Mall et al., 2015; Ragupathy et al., 2008; Singh et al 2012; Uprety et al., 2010) also grouped all the ailments reported by ethnic groups in to different disease categories and calculated Fic values. Fic value indicates the agreement or disagreement in the use of taxa for treatments of the diseases among the tribal.

The highest Fic values for muscular/skeletal disease category in the present study indicates a remarkable agreement of the informants regarding the use of plants for treatment of muscular sprain and bone fracture in the study area. The Dura community valued four plant species -*Calotropis gigantea*, *Drynaria quercifolia* (L.) J. Sm., *Hoya lanceolata* Wall. ex D. Don, and *Periploca graeca* L. - for their cultural significance in treating muscle sprains and bone fractures. The factor of informant consensus provides a measure of reliability supporting the claims regarding the use of plants for medicinal purposes in the ethnomedicinal studies (Malla et al., 2015). A higher level of consensus about the use of particular taxa for curing ailments indicates that the ethnomedicinal use of plants is in practice (Shrestha et al., 2014; Singh et al., 2012).

The Comparatively low consensus factor (Fic 0.85) for the eye problem category may indicate a lower prevalence of eye problem among the Duras. The data also showed that use report for eye and kidney problem were comparatively low. Fic values not only reflect agreement regarding the use of taxa for the treatment of disease, but also indicate the use reports of the taxa (Ragupathy et al., 2008).

Table 1: FIC value for different disease categories

S.N.	Disease category	Nt	Nur	Fic
1	Gastrointestinal	47	988	0.95
2	Respiratory	27	531	0.95
3	Dermatological	20	477	0.96
4	Muscular/ Skeletal	4	104	0.97
5	UTI	5	58	0.92
6	Pain	25	462	0.94
7	Kidney stone	3	16	0.87
8	Eye problems	4	21	0.85
9	Jaundice	7	77	0.92
10	Blood related problems	16	236	0.93
11	Other	41	673	0.94

Among the eleven disease categories, the muscular/ skeletal disease category had the highest factor for informant consensus (Fic) value. This highest Fic value indicates a remarkable homogeneity of knowledge of the informant regarding the use of plants for treating muscular sprain and bone fracture in the study area. The Dura tribe considered four plant species -*Calotropis gigantea*, *Drynaria quercifolia*, *Hoya lanceolata*, and *Periploca graeca* - culturally significant for treating these conditions. Phytochemical test of culturally valued plants among the Dura community must be done to validate the knowledge scientifically. Specifically, extraction and phytochemical analysis of these four plant species should be done to test for the presence of anti-inflammatory compounds, as anti-inflammation is key principle in the treatment of fractures.

Conclusion

A total of 73 medicinal plant species used by the Dura people to treat 64 different ailments, were recorded. The Duras have native names for these plant species, many of which differ significantly from those in the Gurung language. For some species, the Duras use Nepali names. Asteraceae was the most prominent family followed by Lamiaceae, Moraceae, Apocynaceae, and others. Herbs were the dominant form among the medicinal plants, and leaves were the most frequently used plant part. Much frequent used form of medicine was plant juice, likely due to the easily availability of these plants.

From this study, it can be concluded that although the Dura are a marginalized tribe with small population, they have their own language, primary healthcare culture, and deep knowledge on medicinal plants and the ailments they commonly face. Their use of medicinal plants for primary health care remains high. The Dura tribe is one of the heritages of Nepal, so we should focus to preserve the various aspects of their traditional knowledge and practices.

Author Contributions

Both authors were involved in the research. A Ghimire visited the study site, collected ethno-medicinal data and plant specimens, and prepared the herberium and C M Nemkul reviewed the manuscript.

Acknowledgements

We are very thankful to local villagers, healers and informants of Madhyanepal municipality of Lamjung District for their kind cooperation.

References

Acharya, R. (2012). Ethnobotanical study of medicinal plants of Resunga hill used by Magar community of Bada Gaun VDC, Gulmi district, Nepal. *Scientific World, 10*(10).

Adhikari, R. K. (1973-74). The Duras of West Nepal. *Nepal Digest, 2*(11), 59-60.

Bastakoti, N. D. (2019). An ethnobotanical study of medicinal plants used by Kumal and Gandarva community of Pokhara Metropolis, Kaski, Nepal. *Himalayan Biodiversity, 7*, 19-31. https://doi.org/10.3126/hebids.v7i1.40186

Bhandari, R., Pandeya, B., & Ghimire, B. (2023). Ethnobotanical study of plant resources in Dhurkot rural municipality, Gulmi district, Nepal. *Ethnobotany Research and Applications, 25*, 1-19. https://ethnobotanyjournal.org/era/index.php/era/article/view/4301

Bhattarai, K. R. (2017). Ethnomedicinal practices of the Lepcha community in Ilam, East Nepal. *Journal of Plant Resources, 15*(1), 31-44.

Bhattarai, N. K. (1989). Traditional phytotherapy among the Sherpas of Helambu, Central Nepal. *Journal of Ethnopharmocology, 27*(172), 45-54.

Tamang, R., Thakur, C., Koirala, D., & Chapagain, N. (2017). Ethno-medicinal plants used by Chepang community in Nepal. *Journal of Plant Resources, 14*(1), 21-30.

Coburn, B. (1984). Some native medicinal plants of Western Gurungs. *Kailash, 11*, 55-87.

Dangol, D. R., & Gurung, S. B. (1991). Ethnobotany of the Tharu tribe of Chitwan district, Nepal. *International Journal of Pharmacognosy, 29*(3), 203-209.

Frei, B., Baltisberger, M., Sticher, O., & Heinrich, M. (1998). Medical ethnobotany of the Zapotecs of the Isthmus-Sierra (Oaxaca, Mexico): Documentation and assessment of indigenous uses. *Journal of Ethnopharmacology, 62*(2), 149-165

Gebeyehu, G., Asfaw, Z., Enyew, A., & Raja, N. (2014). Ethnobotanical study of traditional medicinal plants and their conservation status in Mecha Wereda West Gojjam Zone of Ethiopia. *International Journal of Pharm Health Care Res, 2*(3), 137-154.

Ghimire, K., & Bastakoti, R. R. (2009). Ethnomedicinal knowledge and health care practices among the Tharus of Nawalparasi district in Central Nepal. *Forest Ecology and Management, 257*, 2066-2072. http://doi.org/10.1016/j.foreco.2009.01.039

Heinrich, M., Ankli, A., Frei, B., Wiemann, C., & Sticher, O. (1998). Medicinal plants in Mexico: Healer's consensus and cultural importance. *Social Science and Medicine, 47*, 1859-1871.

Limbu, D. K., & Rai, B. K. (2013). Ethno-Medicinal practices among the Limbu community in Limbuwan, Eastern Nepal. *Global Journal of Human Social Science, 2*, 7-29.

Luitel, D. R., Rokaya, M. B., Timsina, B., & Münzbergová, Z. (2014). Medicinal plants used by the Tamang community in the Makawanpur district of Central Nepal. *Journal of Ethnobiology and Ethnomedicine, 10*(5).

Malla, B., Gauchan, D. P., & Chhetri, R. B. (2015). An ethnobotanical study of medicinal plants used by ethnic people in Parbat district of Western Nepal. *Journal of Ethnopharmacology, 165*, 103-117. https://doi.org/10.1016/j.jep.2014.12.057

Manandhar, N.P. (1985). Ethnobotanical notes on certain medicinal plants used by Tharus of Dang-Deokhuri district, Nepal. *International Journal of Crude Drug Research, 23*(4), 153-159.

Manandhar, N. P. (1987). Traditional medicinal plants used by tribals of Lamjung District, Nepal. *International Journal of crud Drug Research, 25*(4), 236-240. https://doi.org/10.3109/13880208709055200

Manandhar, N.P. (1991). Medicinal plant lore of Tamang tribe of Kabhrepalanchok district, Nepal. *Economic botany, 45*(1), 58-71.

Manandhar, N. P. (1998). Native phytotherapy among Raute tribes of Dadeldhura district, Far-western Nepal. *Journal of Ethno-pharmacology, 60*, 199-206.

Manandhar, N. P. (2002). *Plants and people of Nepal*. Timber press..

Nemkul, C. M., Bajracharya. G. B., & Shrestha, I. (2018). Ethno-medico botanical Study of Magar community in Dhaubadi VDC, Nawalparasi district, Nepal. In A. Rab, M. Rahman, A. H. Chowdhury, & M. F. Hossain (Eds.), *13th International Knowledge Globalization Conference proceeding* (pp. 55-65). IUBAT-International University of Business, Agriculture and Technology.

Nemkul, C. M., Bajracharya, G. B., & Shrestha, I. (2022). Assessment of antimicrobial and antioxidant activities of four ethnomedicinal plants used by Magars in Nawalpur district, Nepal. *Journal of Plant Resources, 20*(2), 146-155. https://doi.org/10.3126/bdpr.v20i2.57030

Nemkul, C.M. (2022). *Ethnobotany and antimicrobial study of selected medicinal plants used by Magar community in Nawalpur, district, Nepal*. (Unpublished doctoral dissertation). Tribhuvan University.

National Population & Housing Census. (2021). *National Population & Housing Census 2021*. National Statistics Office.

Poudel, S., & Gautam, C. (2008). Ethnomedicine in Magar community of Dhadhing district, Central Nepal. *Bulletin of the Department of Plant Resources, 30*, 80-86. https://www.nepjol.info/index.php/BDPR/article/view/1531.

Ragupathy, S., Steven, N. G., Maruthakkutti, M., Velusamy, B., & Ul-Huda, M. M. (2008). Consensus of the 'Malasars' traditional aboriginal knowledge of medicinal plants in the Velliangiri holy hills, India. *Journal of Ethnobiology and Ethnomedicine*, *4*, 1-14.

Rijal, A. (2011). Surviving on knowledge: Ethnobotany of Chepang community from mid- hills of Nepal. *Ethnobotany Research & Applications*, *9*, 181-215.

Sacherer, J. (1979). The high altitude ethnobotany of the Rolwaling Sherpas. *Contribution to Nepalese Studies*, *4*(2), 45-64.

Shrestha, N., Prasai, D., Shrestha, K. K., Shrestha, S., & Zhang, X. C. (2014). Ethnomedicinal practices in the highlands of central Nepal: A case study of Syaphru and Langtang village in Rasuwa district. *Journal of Ethnopharmacology, 155*(2), 1204-1213. https://doi.org/10.1016/j.jep.2014.07.002

Singh, A. G., Kumar, A., & Tewari, D. D. (2012). An ethnobotanical survey of medicinal plants used in Terai forest of western Nepal. *Journal of Ethnobiology and Ethnomedicine, 8*, 1-15. https://doi.org/10.1186/1746-4269-8-19

Thapa, S. (2012). Medico-ethnobotany of Magar communit in Salija VDC of Parbat district, Central Nepal. *Our Nature*, *10*, 176-190.

Uprety, Y., Asselin, H., Boon, E. K., Yadav, S., & Shrestha, K. K. (2010). Indigenous use and bio-efficacy of medicinal plants in the Rasuwa district, Central Nepal. *Journal of ethnobiology and ethnomedicine*, *6*, 1-10.

Appendix 1: Ethnomedicinal data

Family	Botanicalname, Voucher no.	Name in DURA language	Local name	Habit	Type	Ailments	Parts used	Drug forms
Acanthaceae	*Justicia adhatoda* L. Ga27	Asuro	Asuro	Shrub	Wild	Tonsillitis, cough, asthma	Root, Flower, Young leaf	Powder, Decoction
Acoraceae	*Acorus calamus* L. Ga 28	Syade	Bojho	Herb	Cultivated	Sore throat, cough and cold, paralysis, epilepsy	Root, Rhizome	Chewing, Paste, infusion
						Anthelmintic in cattle		Juice
Asparagaceae	*Agave americana* L.	Ketuki	Ketuki	Shrub	Wild	Burn, cuts, cancer, indigestion	Leaf	Juice
Amaranthaceae	*Acyranthes bidentate* Blume	Chyuna	Datiwan	Herb	Wild	Toothache, skin diseases, gastritis, hair fall	Root	Juice
							Stem	Chewing
	Alternanthera sessilis (L.) R. Br. ex DC.	Bhringi raj	Bhringi jhar	Herb	Wild	Asthma, cut wounds, fever	Leaf	Juice and Paste
Anacardiaceae	*Magnifera indica* L.	Aanbo	Aap	Tree	Cultivated	diarrhea, menstrual problem, ulcer, asthma	Bark	Decoction
						Bee sting	Sap	Apply sap
	Rhus chinensis Mill. Ga 29	Bhakimlo	Bhakimlo	Shrub	Wild	Diarrhea, blood purification, stomach pain	Fruit, Leaf	Decoction
Apiaceae	*Centella asiatica* (L.) Urb. Ga40	Ghodtapre	Ghodtapre	Herb	Wild	Cut wounds, diarrhea, fever, sore throat, indigestion, ulcer, eye problems	Whole plant	Paste Juice
						Urine infection (blockage) in cattle		Juice
Apocynaceae	*Alstonia scholoris* (L.) R.Br. Ga30	Chhatiwan	Chhatiwan	Tree	Wild	Ulcer, dysentery, malaria, skin diseases in both cattle and human, lactation	Bark, Leaf	Powder, Decoction, Juice
	Calotropis gigantean (L.) Dryand	Aank	Aank	Shrub	Wild	Gastritis, paralysis, body pain, wounds, sprain, fracture	flower, sap, root, leaf	Paste, Decoction
	Hoya lanceolata Wall. ex D. Don Ga31	Thirjo	Thirjo	Shrub	Wild	Back pain, fracture, sprain	Whole plant	Decoction
	Periploca graeca. L. Ga 13	Shikhari lahara	Shikari lahara	Climber	Wild	Fracture, joints pain, diabetes, Gastritis, high blood pressure	Whole plant	Chewing, Paste
						Wounds in cattle due to tiger/ lion attack		Paste
Asperagaceae	*Asparagus racemosus* Willd. Ga39	Kurilo	Kurilo	Shrub	Cultivated	Dysentery, common cold, jaundice, menstrual problems, lactation in both human and cattle	Leaf, Root, rhizome	Decoction
Asphodelaceae	*Aloe vera* (L.) Burm.f.	Doma	Ghiukumari	Herb	Cultivated	Burn, high blood pressure, diabetes, acne, pimples, gastritis	Leaf	Apply gel, Consume Juice or gel
Asteraceae	*Acmella oleracea* R.K. Jansen	Phaka	Marati	Herb	Cultivated	Indigestion, anthelmintic	Fruit	Paste
	Ageratina adenophora (Spreng.)	Banmasa	Banmara	Shrub	Wild	Cuts and wounds, scabies, head	Leaf,	Juice, Paste,

Family	Botanicalname, Voucher no.	Name in DURA language	Local name	Habit	Type	Ailments	Parts used	Drug forms
	R. M. King & H. Rob.					ache, sinusitis	Young stem, Root	inhale vapor
	Artemisia vulgaris L.	Paati	Titepati	Herb	Wild	Anthelmintic, gout, skin diseases, scabies	Leaf	Paste, Juice
	Elephantopus scaber L.	Sahasra buti	Sahasra buti	Herb	Wild	Common cold, tuberculosis, high fever	Root	Decoction
						Abdominal bloating in cattle	Whole plant	Decoction, Juice
	Galinsoga parviflora Cav.	Gande	Gandhe	Herb	Wild	Cuts and wounds, diarrhea	Leaf	Juice, Paste
	Inula cappa (Buch.-Ham. ex D.Don) DC	Jyori	Gaitihare	Herb	Wild	Rheumatoid arthritis, body pain	Root, Leaf	Decoction
Berberidaceae	*Berberis asiatica*Roxb. Ex DC. Ga38	Chutro	Chutro	Shrub	Wild	Piles, ye problem, jaundice, fever	Root, stem, Bark, Fruit	Paste, Powder, Decoction, infusion
Cannabaceae	*Cannabis sativa* L. Ga37	Dhaso	Ganja	Herb	Cultivated	Common cold, dysentery, abdominal bloating in cattle	Leaf, Seed	Powder
Capparaceae	*Crataeva magna* (Lour.) DC.	Sipliyan	Sipligan	Tree	Cultivated	Kidney stone, fever, urine infection	Leaf, Stem	Decoction
Caryophyllaceae	*Drymaria cordata* (L.) Wild. Ex schult Ga36	Abijalo	Abhijalo	Herb	Wild	Common cold, gastritis, nausea	Whole plant	Paste and juice
Combretaceae	*Terminalia bellirica* (Gaertn.) Roxb. Ga 15	Thechu	Barro	Tree	Wild	Piles, gastritis, cough, common cold	Fruit	Powder, Decoction
	Terminalia chebula Retz. Ga 14	Harro	Harro	Tree	Wild	Gastritis, ulcer, menstrual problems	Fruit	Chewing, powder, Decoction
Costaceae	*Costus speciosus* (Koen ex. Retz.) Sm.	Betbara	Betlauri	Herb	Cultivated	Snake bite, stomach pain, gastritis	Root, rhizome	Paste, powder
Crassulaceae	*Kalanchoe pinnata* (Lam.) Pers.	Kle	Ajambari jhar	Herb	Wild	Jaundice, kidney stone, gastritis, dyspepsia	Leaf	Juice
Ericaceae	*Rhododendron arboreum* Sm.	Bha	Lali gurans	Tree	Wild	Diabetes, blood pressure, bone stuck in throat	Flower	Chewing and Juice
Euphorbiaceae	*Jatropha curcas* L.	Sajiwan	Sajiyon	Shrub	Wild	Toothache, tonsillitis, burn	Leaf, stem	Juice and Paste
Fabaceae	*Bauhinia variegate* (L.) Ga35	Koiralo	Koiralo	Tree	Cultivated	Blood cancer, liver diseases, diarrhea, piles, menstrual problem	Bark, Flower, Leaf	Juice and Decoction
	Caesalpinia decapetala (Roth) Alston	Areli	Areli	Shrub	Wild	Stomach pain, diarrhea	Root	Chewing
						Mastitis in cattle	Stem, leaf	Roasting
	Mimosa pudica L. Ga34	Buhari jhar	Lajjawati jhar	Herb	Wild	Piles, fever, kidney stone, urine infection, gastritis, lactation	Leaf, Root	Paste, hewing, Powder
Fagaceae	*Castanopsis indica* (Roxburgh ex Lindil.) A. DC.	Kadush	Katush	Tree	Wild	Indigestion, joints pain	Bark	Decoction
Lamiaceae	*Callicarpa macrophylaa* Vahl. Ga 26	Dahikamala	Dahikaamlo	Shrub	Wild	Sore throat, ulcer, skin diseases	Root, fruit	Powder

Family	Botanicalname, Voucher no.	Name in DURA language	Local name	Habit	Type	Ailments	Parts used	Drug forms
	Leucosceptrum canum Sm.	Dhursul	Dhursyauli	Shrub	Wild	Headache, fever	Leaf, Root	Decoction
	Mentha spicata L. Ga25	Binidzu	Pudina	Herb	Cultivated	Nausea, gastritis, fever, diarrhea	Leaf	Paste and Decoction
	Ocimum basilicum L. Ga24	Babari	Babari	Shrub	Cultivated	Urine infection, headache, eye problem	Root, Leaf, flower, Bark	Powder, Decoction, Juice
	Ocimum tenuiflorum L. Ga 23	Tulasi	Tulasi	Shrub	Cultivated	Fever, cough and cold, diabetes, gastritis, heart disease, ulcer	Leaf, Root, Stem	Chewing, Decoction, Juice
Lythraceae	*Woodfordia fruticosa* (L.) Kurz. Ga33	Budho Dangero	Bot Dhaiyanro	Herb	Wild	Dysentery, gastritis	Leaf	Powder
Malvaceae	*Bombax ceiba* L.	Eku	Simal	Tree	Wild	Blood purification, stomach pain, wounds	Bark, Sap, Flower, leaf	Decoction
Melastomataceae	*Osbeckia stellata* Buchanan-Hamilton ex D. Don	Angeri	Sano Angeri	Shrub	Wild	Indigestion, toothache, diarrhea	Root, fruit, Leaf	Juice, Decoction
Meliaceae	*Azadirachta indica* A. Juss Ga32	Nim	Neem	Tree	Cultivated	Skin diseases, fever, diabetes, uric acid, high blood pressure	Leaf, Bark	Paste, Juice, powder
Menispermacae	*Cissampelos pareira* L.	Guargano	Gudargano	Climber	Wild	Cough, gastritis, indigestion, fever, diarrhea	Whole plant	Juice and paste
	Stephania japonica (Thunb.) Miers Ga22	Chillo Batulpate	Chillo Badallpate	Climber	Wild	Gastritis, headache	Stem, leaf	Juice
	Tinospora sinensis (Lour.) Merr. Ga31	Tigi	Gurjo	Climber	Wild	Chronic fever, diabetes, jaundice, urine infection, posterior nosebleed	Leaf and Stem	Decoction
Moraceae	*Ficus benghalensis* L.,	Bar	Bar	Tree	Wild	Diabetes, diarrhea, cough and cold, eye problems	Bark, Seed	Decoction, Juice, infusion
	Ficus benjamnia L.	Shami	Shami	Tree	Wild	Chest pain	Fruit	Powder
	Ficus semicordata Buch.-Ham. ex Sm.	Khadayo	Khanayo	Tree	Cultivated	Scabies	Latex	Apply latex
						Headache, indigestion	Fruit, Bark,	Juice
						Miscarriage/ Problem in postpartum discharge in cattle	Bark	Decoction
	Morus alba L.	Kimbu	Kiu kafal	Tree	Cultivated	Toothache, dysentery, diabetes, anthelmintic	Bark, Root, Fruit	Juice and Paste
Moringaceae	*Moringa Oleifera* Lam.	Shital chini	Sajiwan	Tree	Cultivated	High blood pressure, blood purification, liver diseases	Leaf, Fruit	Powder
Myricaceae	*Myrica esculenta* Buch.-Ham. ex D. Don	Kafal	Ban Kaphal	Tree	Wild	Asthma, gastritis, anthelmintic, aigh blood pressure	Fruit, chewable bark	Chewing
Myrtaceae	*Psidium guajava* L.	Basa	Belauti	Tree	Cultivated	Diarrhea, ulcer, gastritis, nausea, toothache	Bark, Fruit, Young leaf	Juice, Decoction
Nephrolepidaceae	*Nephrolepis cordifolia* (L.) C. Presl	Naisu	Pani amala	Fern	Wild	jaundice, blood pressure, indigestion	Fruit	Chewing
Oleaceae	*Nyctanthes arbor-tristis* L.	Shringar	Parijat	Tree	Cultivated	Cough, fever, asthma	Leaf, Bark,	Decoction

Family	Botanicalname, Voucher no.	Name in DURA language	Local name	Habit	Type	Ailments	Parts used	Drug forms
		phul					Flower	
Oxalidaceae	*Oxalis corniculata* L. Ga21	Amilo jhar	Chari amilo	Herb	Wild	Fever, common cold, gastritis, blood purification	Whole plant	Chewing, Juice
Phyllanthaceae	*Phyllanthus emblica* L.	Kon	Amala	Tree	Cultivated	Gastritis, diarrhea, common cold, hair fall, abdominal bloating	Root, Leaf, fruit, Bark	Chewing, juice, decoction
Piperaceae	*Piper longum* L.	Gumthi	Pipla	Climber	Wild	Gastritis, common cold, fever, diabetes	Fruit, Root	Powder
	Piper Nigrum L.	Marich	Gol marich	Climber	Cultivated	Asthma, headache, stomach pain	Seed	Decoction
Poaceae	*Cynodon dactylon* (L.) Pers. Ga 20	Dubo	Dubo	Herb	Wild	Burn, cuts, wounds, scabies, vitiligo	Whole plant	Paste, Juice
Polypodiaceae	*Drynaria quercifolia* (L.) J. Sm. Ga 19	Kamari	Kammari	Fern	Wild	Fracture, Joints pain, Sprain	rhizome, Sap	Apply paste
Rosaceae	*Rubus ellipticus* Sm.	Tanchi	Ainselu	Shrub	Wild	Anaemia, indigestion, low blood pressure, cough	Fruit, Root	Chewing
Rubiaceae	*Mussaenda macrophylla* Wall.	Dhobeni	Dhobeni	Shrub	Wild	Indigestion, gastritis	Root	Juice
Rutaceae	*Aegle marmelos* (L.) Correa. Ga 15	Bel	Bel	Tree	Wild	Diabetes, gastritis, loss of appetite	Leaf, Bark, Fruit	Decoction
	Citrus aurantiifolia (Christm.) Swingle	Gam	Kagati	Tree	Cultivated	Gastritis, jaundice, blood purification	Fruit	Juice
	Zanthoxylum armatum DC.	Promu	Timur	Shrub	Cultivated	Gastritis, fever, dysentery, toothache, common cold, sinusitis	Fruit	Juice, Decoction, Paste
Saxifragaceae	*Bergenia cilliata* (Haw.) Sternb.	Pakhanbed	Pakhanved	Herb	Cultivated	Cuts, wounds, fever, piles, heart diseases	Leaf, Flower, Root	Paste, Decoction
Sinopteridaceae	*Cheilanthes dalhousiae* Hook. Ga 18	Kali singa	Rani sinka	Herb	Wild	Ulcer, gastritis	Frond	Juice
Solanaceae	*Datura stramonium* L. Ga 16	Dhaturo	Dhaturo	Herb	Cultivated	Cough, gout, asthma, rheumatoid arthritis	Seed, Leaf	Powder
	Nicotiana tabacum L.	Surti	Kacho paat	Herb	Cultivated	Wounds	Leaf	Paste
						Anthelmintic in cattle		Juice
Urticaceae	*Urtica dioica* L. Ga 17	Koke	Sisno	Shrub	Wild	Jaundice, diabetes	Young leaf, Root	Juice, Paste, Decoction
Zingiberaceae	*Curcumal Longa* L.	Ghodzu	Haledo	Herb	Cultivated	Common cold, fever, furuncle	Rhizome	Decoction, paste
	Elettaria cardamomum (L.) Malton	Alaichi	Alaichi	Herb	Cultivated	Snake bite, cough and cold	Fruit	Apply paste, Chewing

Journal of Plant Resources (2025) **23**(1), 91-106
https://doi.org/10.3126/bdpr.v23i1.83357

An Assessment of the Ethnobotanical Knowledge of Danuwar Community in Udayapur District, Nepal

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Abstract

This study investigated the Danuwar community's traditional plant-based knowledge dwelling in the Katari municipality, Udayapur district, with the objective of documenting the diversity, medicinal applications, and cultural significance of the plant species they utilize. Data was collected from March to June, 2024 using semi-structured questionnaires, field observations, and group discussions with 89 informants (43 men and 46 women), along with six key informant interviews and two focus group discussions. Data was analyzed in R studio using Ethnobotanical indices: Use value (UV), Use Reports (UR), Relative Frequency of Citation (RFC) and Cultural Value (CV). A total of 111 plant species from 103 genera and 58 families were documented, primarily for medicinal, cultural, and daily use, with herbs being the most commonly used and 24% of documented species were used to treat gastro-intestinal/digestive disorders. The most significant species was *Achyranthes aspera* (RFC 0.568) whereas *Aegle marmelos* had the highest use value (UV 0.784) with 69 use reports. Similarly, *Achyranthes aspera* shows culturally significant, with their highest Cultural Value (CV 0.100). Deforestation, encroachment, grazing, forest fire and unsustainable practices threatened the use of these valuable plants. Raising awareness about the importance of non-timber forest products and empowering the Danuwar community can help to preserve their cultural heritage and medicinal practices for future generations.

Keywords: Cultural value, ethnobotany, Medicinal plants, Traditional knowledge

Introduction

Ethnobotany is the study of the relationship between plants and people: From "ethno"-study of people and "botany"-study of plants (Rahman et al., 2010). The term "ethnobotany" was first given by John Hershbergerin 1896 which deals with the science of documentation and conservation of original knowledge, which has been used by ethnic people since ancient history (Manandhar, 2002). Ethnobotany focuses on how plants have been utilized, managed and perceived in human societies which includes plants for food, medicine, divination, cosmetics, dyeing, textiles, for building, tools, currency, clothing, rituals and social life (Rahman et al., 2010).The documentation of age-old knowledge plays a vital role in safeguarding biodiversity and responsibly handling natural resources.

Though the country covers only 0.1% of the Earth's terrestrial territory, it is home to about 2% of global biodiversity and ranks 25th in terms of species richness (Kunwar et al., 2021). Nepal is the country with the most diverse and superb landscape with a diversity of plants, animals and fungi (Pangeni et al., 2020), due to its significant variations in altitude, topography and climate, Nepal has an important floral biodiversity comprising nearly 6500 species of flowering plants, (Shrestha et al., 2018) of which about 1,600 to 1,900 species are commonly used in traditional healing practices (Gaire & Subedi, 2011).

In Nepalese communities, medicinal plants are an integral part of traditional medicine (Bhattarai, 2018). They are used as basic ingredients in Ayurveda, Tibetan, homeopathic, and mainstream medicine, among other medical practices (Ghimire et al., 1999). A significant majority, over 75% of the Nepalese population, still relies on indigenous herbal remedies for healthcare due to limited access to modern medical services in rural areas and a shortage of trained healthcare professionals (Kafle & Dani, 2022). There have been more than 590 ethnobotanical studies carried out in Nepal (Joshi & Joshi, 2005; Shrestha et al., 2004). A review of 822 publications made between 1968 and 2014 was done by Rajbhandary and Winkler (2015) which revealed that 44% of published studies were associated with ethnomedicine and 23% with ethnobotany.

Nepal consists of a diverse population of 29.22 million people, consisting of 142 different ethnic groups speaking 124 distinct languages (National Population & Housing Census [NPHC], 2021). Indigenous knowledge (IK) is abundant among the ethnic groups and is firmly established in their traditions and cultures. Danuwar, an indigenous tribe of Nepal comprising population of 82,784 (NPHC, 2021), has a unique culture, traditions, language, and history. The word 'Danuwar' is derived from the Sanskrit word Droniwar. In Sanskrit, 'droni' means flat land between two hills. The people who lived in the inner Tarai between the mountains were called Droniwar and hence, they are called Danuwar in Nepali (Regmi & Thakur, 2016). Danuwars are found throughout the hills and Tarai regions of central and eastern Nepal but they mainly reside in Sindhuli and Udayapur district. There are mainly three clans in Danuwar: Janghariya, Rai Danuwar, and Kachhadiya Danuwar (Bhandari, 1989). Katari Municipality consists of Rai and Kachhadiya Danuwar speaking Nepali and Maithili language respectively (Regmi & Thakur, 2016). They have extensive knowledge about plant utilization and medicinal practices. Agriculture is the Danuwar's primary occupation. Although fishing and hunting were the Danuwar people's traditional occupations, farming is now their main source of income (Acharya & Bhandari, 2024). Limited research exists on Danuwar in Nepal. Furthermore, Katari Municipality of Udayapur district remains unexplored till date on the current status of ethnomedicinal practices and the conservation of plants. Large number of medicinal plants in the region along with their indigenous knowledge is still waiting for proper documentation, thus, making this study vital for documenting traditional knowledge and preserving indigenous culture.

Materials and Method

Study area

Udayapur, Inner Tarai district situated in the Eastern part of Nepal in Koshi Province, lies between (26°392 -27°222 N latitude and 86°92 -87°102 E longitudes). Its geographical limit is 2,063 sq. km. and altitudinal variation starts from 90 m to 2340 m above the sea level. The area has tropical and subtropical climate with an annual minimum temperature of 16.8°C, and an annual maximum temperature of 28.1°C and annual rainfall is about 1349.2 mm. Due to the unique geographical features and various climatic conditions (from lower tropical to temperate), the area is endowed with excellent habitats for diverse flora and fauna. Different topography, geology, and altitude have established three distinct physiographic zones i.e., Inner Tarai, Churia and Mahabharat range. Katari one of the municipality of the Udayapur district consists the population of 59,507 and 20,240 households (NPHC, 2021). Out of 82,784 Danuwar population in Nepal, 9.3% lives in the Katari Municipality (NPHC, 2021). The present research was mainly focused on one ward i.e. ward no. 4 of Katari Municipality which consists an area of 15.46 sq. km. and Danuwar households of about 250.

Data collection

Ethnobotanical data was collected from March to June 2024 through field observations and semi-structured questionnaires administrated to 89 informants (43 men and 46 women) belonging to the Danuwar ethnic group. Additionally, six key informant interviews and two focus group discussionswas conducted with traditional healers, elderly individuals, and frequent users of medicinal plants. The Danuwar ethnic group was selected purposively, while individual respondents within the community were selected randomly. Herbarium specimens were collected during field visits and identified using standard taxonomic reference (Shrestha et al., 2018) with plant nomenclature following the guidelines of Shrestha et al. (2018). Voucher specimens numbered *P. Sharma 01*(FOF) to *P. Sharma 19* (FOF) were deposited at the Faculty of Forestry Herbarium, Hetauda and their identification was cross-verified with existing collections at the National Herbarium and Plant Laboratories (KATH), Kathmandu. All research activities were conducted in accordance with the Code of Ethics of the International Society of Ethnobiology

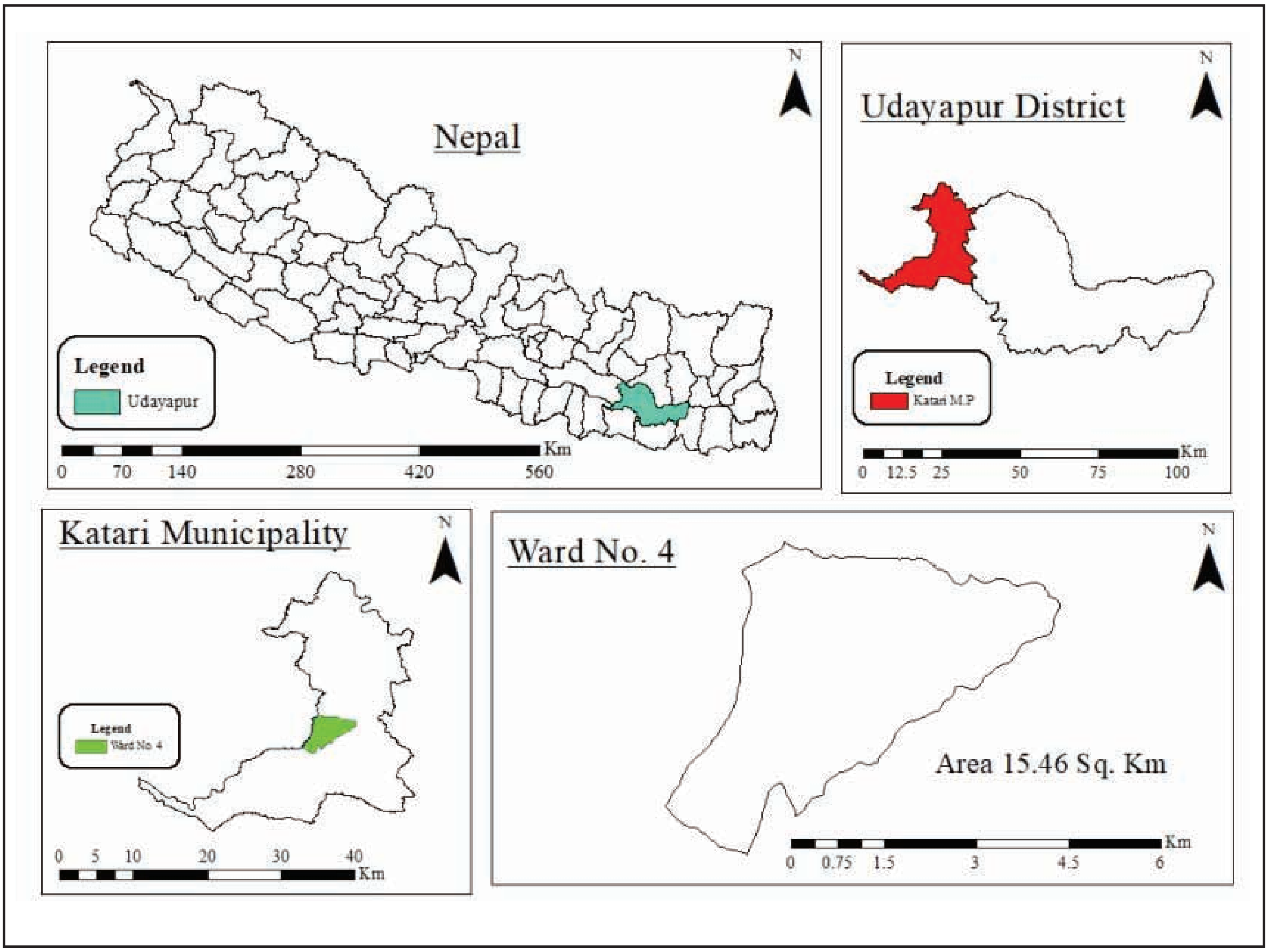

Figure 1: Map of study area

(http://ethnobiology.net/code-of-ethics/). Prior to participation, verbal informed consent was obtained from each respondent through a process of free, prior, and informed consent. Participants were fully informed about the research objectives, their voluntary participation, their right to withdraw at any time, and the measures taken to ensure anonymity and confidentiality.

Data analysis

MS-Excel was used to assess life form; plant parts used, mode of preparation and R studio was used to analyze the quantitative indices.

Use Report (URs): The use report URs () function calculates the use report (UR) for each species in the data set. URs= $\sum_{u=u_1}^{u_{NC}} \sum_{i=i_1}^{i_N} UR_{ui}$

URs computes the total uses for a species as reported by all informants (from i1 to iN) within each specific use category. It counts how many informants mention each use category (NC) for that species and sums all reported uses within those categories (from u1 to uNC) (Prance et al. 1987).

Relative Frequency of Citation (RFC): The Relative Frequency of Citation (RFC) is a quantitative measure used to determine the local importance or popularity of a particular plant species (or item of cultural interest) based on how frequently it is mentioned by informants in a study. RFC is obtained by dividing the number of informants who mention the use of the species, also known as frequency of citation (FC), by the number of informants participating in the survey (N).

RFCs=FCs/N

FCs is the frequency of citation for each species s, and N is the total number of informants 9 Interviewed in the survey (Tardío & Pardo-De-Santayana, 2008).

RFC values range from 0 to 1. A higher RFC means the plant is widely recognized and used among the informants, indicating higher local importance.

Use Value (UV): The relative importance of ethno medicinal plant species was calculated by using the use value (UV) for each species (Phillips et al., 1994).

UVs= $\sum \frac{Us}{Ns}$

Where Us = Total number of use-reports cited by each informant for a given plant species s and Ns = Total number of informants interviewed for plant species.

Cultural Value (CV): Cultural Value (CV) is a comprehensive index that reflects the overall significance of a plant (or other cultural item) to a community. It considers not only how frequently a plant is cited but also how many different uses it has and how widely these uses are known among the population.

CV=$\left[\frac{NU_s}{NC}\right] \times \left[\frac{FC_s}{N}\right] \times \left[\sum_{u=u_i}^{u_{NC}} \sum_{i=i_1}^{i_N} UR_{ui}/_{N}\right]$

Where, NUs = No of uses, NC = No. of Use Categories, FCs = Frequency of Citation, N = No. of Informants (Reyes-García et al., 2006).

A higher CV indicates a plant with many uses, known by many people, and of high cultural importance.

Results and Discussion

Plant diversity and uses

From the present study 111 plant species were recorded as ethnobotanical use, and they belonged to 58 families and 103 genera (Appendix 1). Lamiaceae represented the highest number of plants (11 species), which was followed by Fabaceae (8 species) and Euphorbiaceae (5 species). The herbs were primary sources of medicine (43%), followed by tree (29%) and shrub (18%). The study showed that different parts of the same plants were used for different purposes (food, medicine, cultural). The uses of plant species were classified into 11 categories (traditional medicine, species used to cure animal disorder, vegetables, edible fruits, cultural value, ritual, spice, tonic, NTFPs contributing economy, agricultural food, and plant related with taboos) (Table 1). The highest number of species (105) was used as traditional medicine, followed by species used as vegetables (15) and edible fruits (13). The extensive utilization of medicinal plants indicates inhabitants of the studied areas possess rich traditional knowledge, enabling them to treat various ailments using locally available natural resources. Plants are utilized as food, cultural value, contributing economy, and as a veterinary care in Katari Municipality shows how locals uses the plants for multiple requirements, as also recorded in earlier studies (Manandhar, 2002). The widespread use of these plants was due to easy availability, cultural significance and their ability to cure the illnesses.

Based on the information gathered from informants, the uses of medicinal plants for culinary purpose were categorized into 11 ailments (Table 2). Out of these, digestive system disorders showed the use of highest number of plants (41), followed by skin /integumentary system disorders with the use of 33 plant species and respiratory system disorders with the use of 21 species. The frequent use of plants for gastrointestinal disorders indicates the significance of these ailments and suggests the potential existence of additional, yet undocumented, ethnomedicinal knowledge relevant to the treatment of gastrointestinal conditions. This information is similar to findings of several studies (Bhattarai, 2017; Kafle & Dani, 2022; Shrestha et al., 2014). First choice of majority of people is traditional medicine and the Dhami/Jhankri systems; if that doesn't work, they look for alternative solutions. The villagers treat simple common illnesses including cough and allopathic cold, scrapes and wounds, low fever, typhoid, jaundice, pains, and other conditions at home. This shows that the primary healthcare services used by the locals have not entirely relied on modern medicine.

Table 1: Ethnobotanical category of plant species

S.N.	Categories	Plant species	No. of species
1	Plant-based food (Vegetables)	*Amorphophallus paeoniifolius, Asparagus racemosus, Bambusa bambos, Brassica juncea, Cantharellus odoratus, Celosia argentea, Jatropha curcas, Lablab purpureus, Musa×paradisiaca, Ophioglossum reticulatum, Oroxylum indicum, Oxalis corniculata, Senna tora, Syzygium aromaticum, Urtica dioica*	15
2	Edible Fruits	*Aegle marmelos, Ananas comosus, Areca catechu, Artocarpus lacucha, Carica papaya, Citrus aurantiifolia, Cocos nucifera, Mangifera indica, Melastoma malabathricum, Musa×paradisiaca, Phyllanthus emblica, Syzygium cumini, Ziziphus mauritiana*	13
3	Cultural Value	*Achyranthes aspera,Aegle marmelos, Bambusa bambos, Calotropis gigantea, Dalbergia sissoo, Elsholtzia fruticosa, Ficus religiosa, Mangifera indica, Phyllanthus emblica, Shorea robusta, Tagetes erecta*	11
4	Species used to cure animal disorder	*Allium*sp., *Alstonia scholaris, Artemisia indica, Asparagus racemosus, Cannabis sativa, Cissus quadrangularis, Datura metel, Melia azedarach, Opuntia monacantha, Pogostemon benghalensis, Zingiber cassumunar*	11
5	Ritual	*Areca catechu, Bombax ceiba, Cocos nucifera, Musa× paradisiaca, Ocimum tenuiflorum, Oroxylum indicum, Saccharum officinarum, sesamum indicum, Vitex negundo*,	9
6	Spice	*Capsicum frutescens, Mentha spicata, Perilla frutescens, Sesamum indicum, Syzygium aromaticum*	5
7	Tonic plant	*Brassica campestris, Cocos nucifera, Leucas aspera, Terminalia chebula, Tinospora sinensis*	5
8	NTFPs contributing economy	*Bambusa bambos, Mangifera indica, Phoenix acaulis, Shorea robusta*	4
9	Agricultural Food	*Macrotyloma uniflorum, Oryza sativa*	2
10	Plant related with Taboos	*Cannabis sativa*	1

Table 2: Aliments categories of medicinal plants

S.N.	Categories of ailments	Ailments type/medication	No. of species used
1	Digestive System	Gastritis, Swollen stomach, Mouth infection, Diarrhea, Vomiting, Constipation, Dysentery, Stomach disorder, Food aversion, Fish bone pricking, Yellowish teeth, Indigestion, Appetizer, Stomache, Toothache, Worm infection, Energy boost, Piles.	41
2	Skin and / Integumentary System disorder	Allergy, Bleeding, Boils, Burn, Wounds and cuts, Foot/hand cracks, Scar removing, Scorpion bite, Skin infection, Skin irritation due to latex, Hair tonic, Hair color, Hair growth, Chicken pox, Snake bite, Anti-inflammatory, Trench foot, Dog bite, Remove leech.	33
3	Respiratory System disorder	Asthma, Common cold, Cough, Tonsillitis, Sinusitis, Chest pain.	21
4	Fever		14
5	Domestic animal disorder	Milk production, Bone breakage, Diarrhea, Cut and wounds, Foot and mouth disease, Weight gain, Corneal ulcer.	11
6	Reproductive System disorder	Menstrual cramp, Milk production, Vaginal discharge,	8
7	Skeleto-Muscular system disorder	Body pain, Swollen body, Bone breakage.	8
8	Cardiovascular System disorder	Balance Blood pressure, Blood purification	6
9	Nervous system disorder	Headache, Cooling agent	6
10	Urinary Disorder	Kidney stone, Piles, Urinary troubles,	5
11	Others	Sugar, Jaundice, Typhoid	19

Life form of species, plants parts used and mode of preparation of traditional remedies

From the reported species herbs showed 43% of the use, followed by trees (29%), shrubs (18%) least with climbers (10%) (Figure 2). Dominance of herbs resembled with various other studies from Nepal (Ambu et al., 2020; Luitel et al., 2014; Malla et al., 2015; Rokaya et al., 2010). Herbs were preferred over other types possibly because they are more readily available, easier to gather, and more convenient to carry (Uprety et al., 2010). Different parts of plants-leaves (42), fruits (31), roots (26), bark (14), latex (8), stem (10), seeds (8), flowers (6), whole plant (6) and rhizome (3) were used to cure various ailments and used for various purposes (Figure 3). Because of their easy availability, different aerial parts of medicinal plants, such as leaves, flowers, and fruits were commonly used. Based on the types of plant parts used, various modes of preparation were recorded for treating human ailments. The most common mode of use was plant juice (33), followed by decoctions (18), pastes (15), infusions (10), powders (8), cooked forms (8), massages (6), poultices (7), raw applications (7), topical use (5), and less frequently by fumigation (1), smoke (1), and steam inhalation (1) (Figure 4). The majority of plants were used highly in the form of juice, align to that of Ambu et al. (2020) and Malla et al. (2015). Most remedies were prepared and used immediately after harvesting. Fresh forms were favored since this form is considered to be strong and healthy. The majority of herbal preparations are administered orally to treat human disorders, which have been found to be useful. Similar findings have been documented in number of research related with ethnic groups in Nepal, nearby countries, and globally (Bhattarai et al., 2011; Luitel et al., 2014; Uprety et al., 2010). The collection places of the recorded plant species were mostly from wild (46), followed by cultivated (41), agricultural land (18) and roadside (7). Similar observations were also reported by Malla et al. (2015).

Source of knowledge among various age groups

Traditional healers in the study area claim that their ancestors were the primary source of their traditional

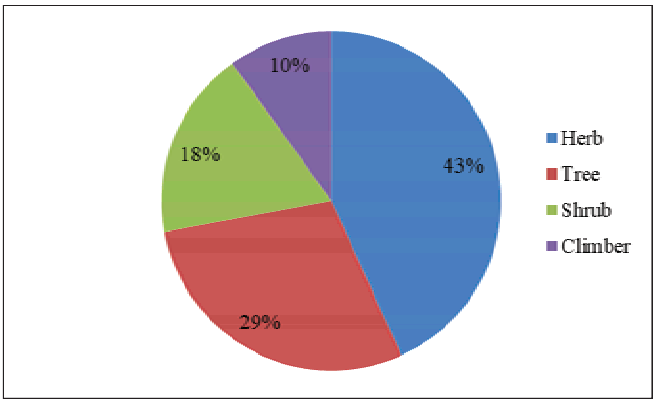

Figure 2: Life form of identified species

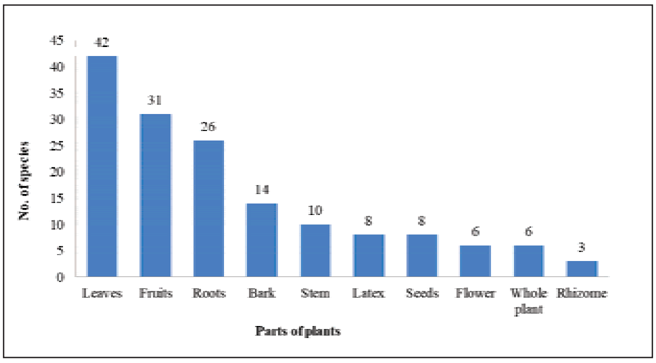

Figure 3: Plants parts used

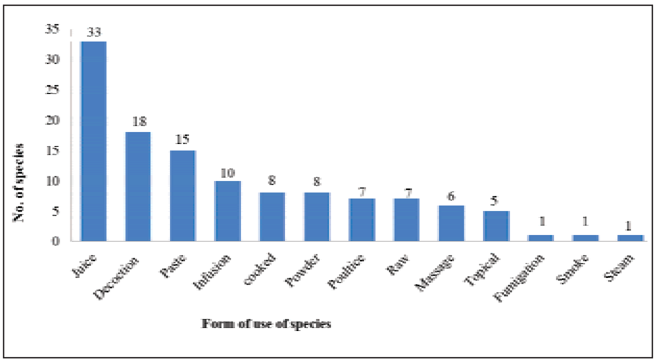

Figure 4: Form of use of species

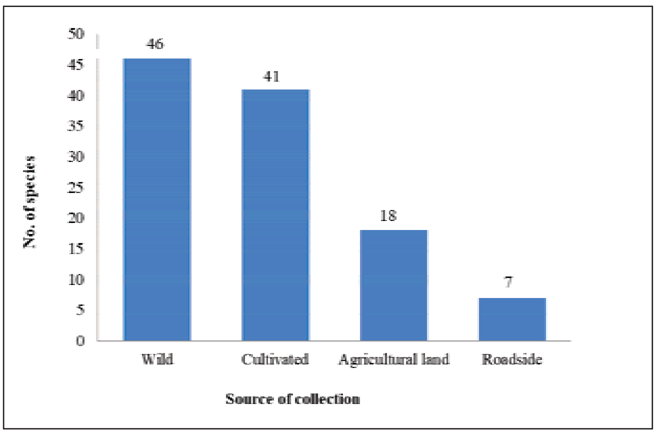

Figure 5: Source of collection of recorded species

knowledge. The other knowledgeable groups, in addition to Dhami/Jhankri, learned about the use of medicinal plant species from family members who had previously used such therapeutic techniques. During the interviews, the elderly people were most familiar with the various types of medicinal plant species. When comparing males and females, females were better at identifying plant species than males because they regularly visit forests for firewood or fodder collection. This information is similar to Luitel et al. (2014) and TorresAvilez et al. (2016). Due to the effects of modernization, fast land degradation, deforestation, and people's reliance on verbal transformation, traditional medical knowledge and practices are rapidly disappearing. Additionally, experts do not transfer or share their knowledge among learners or younger generations. Traditional healers kept their knowledge secret, sharing their knowledge only within their families to preserve efficacy which was also reported by many previous researchers (Bhattarai, 2017; Luitel et al., 2014; Rokaya et al., 2010; Shrestha et al., 2014). It was observed that the poor education level/illiteracy amongst this community led them to be dependent fully on the superstitious culture of Dhami during sickness, relevant to the study by Ambu et al. (2020).

Ethnobotanical indices

According to Use Value (UV), *Aegle marmelos* (L.) Correa (UV=0.784) was the most important with 69 use reports. It was followed by *Bambusa bambos* (L.) Voss (UV=0.682) with 60 use reports and *Oroxylum indicum* (L.) Kurz (UV=0.636) with 56 use reports. The species with least importance were *Ricinus communis* L., *Spondias pinnata* (L.f.) Kurz. and *Syzygium nervosum* DC. with only 3 use reports respectively (Table 3). The highest use value of *Aegle marmelos* (UV=0.784) indicates that it was utilized in a wide variety of contexts, potentially spanning food, medicine, and rituals. The lowest Use Value (UV) reflects lesser recognition or fewer uses reported for the plants. *Achyranthes aspera* L. has the highest Relative Frequency of Citation (RFC=0.568) followed by *Centella asiatica* (L.) Urb. (0.330) and *Azadirachta indica* A. Juss. (0.318) indicating it was frequently cited by informants, due to its prominent role in both medicinal and cultural contexts. Elevated Relative Frequency of Citation (RFC) values indicate a high level of local usage and cultural importance of these species, suggesting that they embody substantial traditional knowledge and should be prioritized for further ethnobotanical investigation. *Achyranthes aspera* shows the highest Cultural Value (CV) which signifies these species shows more than one usage with cultural significance.

Conclusion

The present study reveals a deep connection between the Danuwar community and local plant species, unveiling the significance of 111 species including *Aegle marmelos, Bambusa bambos, Oroxylum indicum,* as the most important which are primarily used for medicine, cultural, and daily use. The high value of quantitative indices suggests that there is

Table 3: Value of Ethnobotanical indices

S.N.	Species name	Use Reports (URs)	Relative Frequency of Citation (RFC)	Use Value (UV)	Cultural Value (CV)
1.	*Achyranthes aspera* L.	54	0.568	0.614	0.100
2.	*Aegle marmelos* (L.) Correa	69	0.273	0.784	0.076
3.	*Azadirachta indica* A.Juss.	36	0.318	0.409	0.028
4.	*Bambusa bambos* (L.) Voss	60	0.170	0.682	0.066
5.	*Cannabis sativa* L.	38	0.148	0.432	0.014
6.	*Centella asiatica* (L.) Urb.	29	0.330	0.330	0.008
7.	*Leucas aspera* (Willd.) Link	26	0.239	0.295	0.010
8.	*Mangifera indica* L.	49	0.148	0.557	0.035
9.	*Mimosa pudica* L.	20	0.216	0.227	0.007
10.	*Oroxylum indicum* (L.) Kurz	56	0.318	0.636	0.058
11.	*Phyllanthus emblica* L.	48	0.205	0.545	0.040
12.	*Piper longum* L.	19	0.205	0.216	0.006
13.	*Shorea robusta* C.F.Gaertn.	48	0.193	0.545	0.045

high potential for further ethnobotanical research and bio-prospecting. Pants of the study area are facing threats from deforestation, habitat loss, and unsustainable harvesting. Thus, empowering both men and women in the Danuwar community can help in preserving cultural heritage and traditional medicinal practices in that community.

Author Contributions

Both authors contributed in analysis, literature review, and concept formulation. P Sharma conducted an in-depth literature review and composed a preliminary draft of the manuscript whereas S Bhattarai analyzed the quantitative indices, revised manuscript and supervised the study. Furthermore, Both authors worked together to refine the content and assure the validity and integrity of the research.

Acknowledgements

First and foremost first author would like to express deepest gratitude to Division Forest Office Udayapur (Triveni) for providing a comfortable working environment. We would like to acknowledge Mr. Amrendra Kumar Yadav for his insightful feedback and suggestions throughout the project. Our sincere thanks also go to Divisional Forest Officer Anirudh Kumar Sah. Additionally, we appreciate the support of the staff at the Division Forest Office in Udayapur (Triveni); their assistance was crucial at various stages of this project. We are particularly thankful to Mr. Deepak Rai, the Chairperson of Kopila Community Forest, for his assistance during data collection. We extend special thanks to the inhabitant of study area who generously shared their time and information during this process. Special thank goes to Ms. Nisha Adhikari, Ms. Sambridhi Timsena and Mr. Dinesh Subedi for their constant support during data collection. Furthermore, grateful to Mr. Prashant Chalise for providing GIS map of the study area.

References

Acharya, U., & Bhandari, R. (2024). Agriculture practices in the Danuwar community of Nuwakot district, Nepal. *International Journal of Agricultural Science and Food Technology*, *10*(2), 107-113.

Ambu, G., Chaudhary, R. P., Mariotti, M., & Cornara, L. (2020). Traditional uses of medicinal plants by ethnic people in the Kavrepalanchok district, Central Nepal. *Plants*, 9(6), 759.

Bhandari, G.P. (1989). *Bhumrasuwa gaun panchayatka Danuwar janjivanko sanskritik pakshya* (*A cultural aspect of Danuwar livelihood of Bhumrasuwa gaun panchayat*) [Master's thesis, Tribhuvan University]. Nepal.

Bhattarai, K. R., Måren, I. E., & Chaudhary, R. P. (2011). Medicinal plant knowledge of the Panchase region in the middle hills of the Nepalese Himalayas. *Banko Janakari*, *21*(2), 31-39.

Bhattarai, K. R. (2017). Ethnomedicinal practices of the Lepcha community in Ilam, East Nepal. *Journal of Plant Resources*, *15*(1), 31-44.

Bhattarai, K. R. (2018). Ethnobotanical study of plants used by Thami community in Ilam district, Eastern Nepal. *Our Nature*, *16*(1).

Gaire, B. P., & Subedi, L. (2011). Medicinal plant diversity and their pharmacological aspects of Nepal Himalayas. *Pharmacognosy Journal*, *3*(25), 6-17. https://doi.org/10.5530/pj.2011.25.2

Ghimire, S. K., Sah, J. P., Shrestha, K. K., & Bajracharya, D. (1999). Ecological study of some high altitude medicinal and aromatic plants in the Gyasumdo valley, Manang, Nepal. *Ecoprint*, *6*(1), 17-25.

Joshi, A. R., & Joshi, K. (2005). *Ethnobotany and conservation of plant diversity in Nepal: Status, bibliography and agenda for sustainable management*. Rubrick.

Kafle, R., & Dani, R. S. (2022). Ethnomedicinal practices of plants in Danuwar community of Dudhauli Municipality, Sindhuli district, Central Nepal. *Journal of Plant Resources*, *20*(2), 211-234. https://doi.org/10.3126/bdpr.v20i2.57041

Kunwar, R. M., Subedi, B. P., Baral, S. R., Maraseni, T., LeBoa, C., Adhikari, Y. P., & Bussmann, R. W. (2021). Ethnobotany of the Himalayas: The Nepal, Bhutanese, and Tibetan Himalayas.

In R. M. Kunwar, H. Sher, & R. W. Bussman (Eds.), *Ethnobotany of the Himalayas* (pp. 1-39). Springer. https://doi.org/10.1007/978-3-030-57408-6_5

Luitel, D. R., Rokaya, M. B., Timsina, B., & Münzbergová, Z. (2014). Medicinal plants used by the Tamang community in the Makawanpur district of Central Nepal. *Journal of Ethnobiology and ethnomedicine*, *10*, 1-11

Malla, B., Gauchan, D. P., & Chhetri, R. B. (2015). An ethnobotanical study of medicinal plants used by ethnic people in Parbat district of Western Nepal. *Journal of Ethnopharmacology, 165*, 103-117. https://doi.org/10.1016/j.jep.2014.12.057

Manandhar, N.P.(2002).*Plants and people of Nepal.* Timber press.

National Population & Housing Census. (2021). *National Population & Housing Census 2021*. National Statistics Office.

Pangeni, B., Bhattarai, S., Paudyal, H., & Chaudhary, R. P. (2020). Ethnobotanical study of Magar ethnic community of Palpa district of Nepal. *Ethnobotany Research and Applications*, *20*, 1-17. https://doi.org/10.32859/era.20.44.1-17

Phillips, O., Gentry, A. H., Reynel, C., Wilkin, P., & Galvez Durand B, C. (1994). Quantitative ethnobotany and amazonian Conservation. *Conservation Biology*, *8*(1), 225-248. https://doi.org/10.1046/j.1523-1739.1994.08010225.x

Prance, G. T., Baleé, W., Boom, B. M., & Carneiro, R. L. (1987). Quantitative ethnobotany and the case for conservation in Ammonia. *Conservation biology*, *1*(4), 296-310.

Rahman, A. H. M. M., Kabir, E. Z. M. F., Sima, S. N., Sultana, R. S., Nasiruddin, M., & Zaman, A. T. M. N. (2010). Study of an ethnobotany at the village Dohanagar, Naogaon. *Journal of Applied Sciences Research, 6*(9), 1466-1473.

Rajbhandary, S., & Winkler, D. (2015). Ethnobotany Nepal: An introduction to the natural history, ecology and human environment of the Himalayas. In G. Miehe, C. A. Pendry, & R. P. Chaudhary (Eds.), *Nepal: An Introduction to the Natural History, Ecology and Human Environment of the Himalayas* (pp. 271-285). Royal Botanic Garden Edinburgh.

Regmi, A., & Thakur, M. I. (2016). *A sociolinguistic survey of Danuwar.* Linguistic Survey of Nepal (LinSuN), Tribhuvan University.

Reyes-García, V., Huanca, T., Vadez, V., Leonard, W., & Wilkie, D. (2006). Cultural, practical, and economic value of wild plants: A quantitative study in the Bolivian Amazon. *Economic botany*, *60*(1), 62-74.

Rokaya, M. B., Münzbergová, Z., & Timsina, B. (2010). Ethnobotanical study of medicinal plants from the Humla district of Western Nepal. *Journal of Ethnopharmacology*, *130*(3), 485-504. https://doi.org/10.1016/j.jep.2010.05.036

Shrestha, K. K., Bhattarai, S., & Bhandari, P. (2018). *Handbook of flowering plants of Nepal (Volume 1. Gymnosperm and Angiosperms: Cycadaceae - Betulaceae).* Scietific Publishers (India).

Shrestha, N., Prasai, D., Shrestha, K. K., Shrestha, S., & Zhang, X. C. (2014). Ethnomedicinal practices in the highlands of Central Nepal: A case study of Syaphru and Langtang village in Rasuwa district. *Journal of Ethnopharmacology*, *155*(2), 1204-1213. https://doi.org/10.1016/j.jep.2014.07.002

Shrestha, K. K., Rajbhandary, S., Tiwari, N. N., Poudel, R. C., & Uprety, Y. (2004). *Ethnobotany in Nepal: Review and perspectives*. WWF Nepal Program.

Tardío, J., & Pardo-de-Santayana, M. (2008). Cultural importance indices: A comparative analysis based on the useful wild plants of Southern Cantabria (Northern Spain). *Economic botany*, *62*, 24-39.

Torres-Avilez, W., Medeiros, P. M. D., & Albuquerque, U. P. (2016). Effect of gender on the knowledge of medicinal plants: Systematic review and meta Analysis. *Evidence Based Complementary and Alternative Medicine, 2016*(1), 6592363.

Uprety, Y., Asselin, H., Boon, E. K., Yadav, S., & Shrestha, K. K. (2010). Indigenous use and bio-efficacy of medicinal plants in the Rasuwa district, Central Nepal. *Journal of ethnobiology and ethnomedicine, 6*, 1-10.

Appendix 1: Identification of plant species with their uses

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Abrus precatorius L.	Fabaceae		Lal Gedi	Herb	fruits	Powder	Agricultural land	Eye impurities	Acharya & Acharya, 2009; Miya et al., 2021
Achyranthes aspera L.	Amaranthaceae	*P. Sharma 01* (FOF)	Datiwan	Herb	Roots, seeds	Juice, Infusion	Roadside	Typhoid, Tonsillitis, Sinusitis, Scorpion bite	Kafle & Dani, 2022; Rijal 2011
Aconitum palmatum (D.Don)	Ranunculaceae		Bikhma	Herb	Roots, Bark	Juice	wild	Vomiting, Diarrhea, Fever, Body pain, Irregular menstrual, Sinusitis	Bhattarai, 2018; Uprety et al., 2010
Acorus calamus L.	Acoraceae		Bojho	Herb	Rhizome	Decoction, Powder, Massage	Cultivated	Cough, wounds, Fever, Body pain, Tonsillitis,	Acharya & Acharya, 2009; Ambu et al., 2020; Bhattarai, 1993; Kafle & Dani, 2022; Poudel & Singh, 2016; Rokaya et al., 2010; Shrestha and Dhillion, 2003; Singh et al., 2012; Uprety et al., 2010
Adina cordifolia (Roxb.) Hook.f. & Benth.	Rubiaceae		Kadam	Tree	Bark, leaves	Paste, Poultice	Wild	Anti-inflammatory , Toothache	
Aegle marmelos (L.) Correa	Rutaceae		Bel	Tree	Fruit, Leaves	Juice	Wild	Cooling agent, Tonsillitis, Blood purification, Edible fruits, Used in Ritual	Ambu et al., 2020; Kafle & Dani, 2022; Panthi & Chaudhary, 2003; Pangeni et al., 2020
Ageratum conyzoides L.	Asteraceae		Gandhe jhar	Herb	Leaves	Paste	Agricultural land	Wounds caused by burns	Singh et al., 2012
Aleuritopteris bicolor (Roxb.) Fraser-Jenk	Pteridaceae		Ratjara	Herb	Root	Infusion	Agricultural land	Fever, Worms infection	Kafle & Dani, 2022
Allium sp.	Amaryllidaceae		Ban pyaj	Herb	Root	Powder	wild	Used during bone fracture in Goat,	
Aloe vera (L.)Burm.f.	Xanthorrhoeaceae		Ghiu Kumari	Herb	Leaves	Gel/topical	Cultivated	Burns, Remove scars, Hair growth, Lower pressure	Bhattarai et al., 2009; Ghimire, 2016; Joshi et al., 2011; Kafle & Dani, 2022; Miya et al., 2021; Poudel & Singh, 2016; Singh, 2020; Singh et al., 2012
Alstonia scholaris (L.)R.Br.	Apocynaceae		Chattiwan	Tree	Bark	Powder	Wild	Tonic,weight gain in animals, Milk production,	
Alternanthera dentata Stuchlík ex R.E.Fr.	Amaranthaceae		Rato phul	Herb	Leaves	paste	Cultivated	Cuts and wounds	
Amorphophallus paeoniifolius (Dennst.) Nicolson	Araceae		Oal	Herb	Leaves, flower	Cooked	Cultivated	Spinach, Swollen body	Kafle & Dani, 2022
Ananas comosus (L). Merr.	Bromeliaceae		Bhuikatahar	Herb	Leaves , Fruit	Steam	Cultivated	Cold and Cough, Edible fruits, Fever	Kafle & Dani, 2022
Areca catechu L.	Arecaceae		Supari	Tree	Fruit	Raw	Cultivated	Scar removing, Beautify the mouth, Induce purification, Edible fruits, Used in Ritual	Kafle & Dani, 2022; Poudel & Singh 2016

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Artemisia indica Willd.	Asteraceae	*P. Sharma 02*(FOF)	Titepati	Herb	Leaves	Paste, Decoction, Infusion	Wild	Allergy, Cuts and Wounds in both Human and Animal, Abdominal pain, Chicken pox,	Kafle & Dani, 2022; Pangeni et al., 2020; Singh et al., 2012
Artocarpus lacucha Buch.-Ham.	Moraceae		Barhar	Tree	Bark, Fruit, leaves	Powder, Infusion	Wild	Pimples, cracked skin, Fever, Edible fruits, Used as Fodder for animals	
Asparagus racemosus Willd.	Asparagaceae		satawari/ kurilo	Shrub	whole plant	Juice, Cooked	Cultivated	Milk production in both Women and Animals, Spinach	Ambu et al., 2020; Bhattarai et al., 2009; Kafle & Dani, 2022; Rjbanshi & Thapa, 2019; Shrestha & Dhillion, 2003; Uprety et al., 2010
Azadirachta indica A.Juss.	Meliaceae		Neem	Tree	Leaves, Bark	Decoction, Poultice	Cultivated	Pesticide in Agriculture, Allergy, Fever, Lower Pressure, Snake bite, Toothache, Toothbrush, Vaginal discharge	Kafle & Dani, 2022; Miya et al., 2021; Singh et al., 2012
Bambusa bambos (L.) Voss	Poaceae		Baans	Herb	Young shoot ,leaves	Cooked	wild	Used as Toothbrush, Young shoot as vegetables, Blood purification, Used as building material, Fodder for Livestock,Used during marriage ceremony and funeral	Kafle & Dani, 2022
Bombax ceiba L.	Malvaceae		Simal	Tree	seed	raw	wild	Seed are edible, Whole plant is used during marriage ceremony and funeral, Root is used as tonic	Singh et al., 2012
Brassica juncea (L.)Czern.	Brassicaceae		Rayo	Herb	leaves	Cooked	Cultivated	Used as spinach	
Brassica campestris L.	Brassicaceae		Tori	Herb	seed	Massage	Cultivated	Used for Oil production, Body pain, Hair tonic	Kafle & Dani, 2022
Calotropis gigantea (L.) W.T. Aiton	Apocynaceae		Aakon	Shrub	Milky latex , Root, leaves	Decoction	Roadside	Latex is used to stop bleeding of wounds, lower pressure, Diabetes, Swollen stomach, Used during marriage ceremony	Miya et al., 2021; Singh et al., 2012
Cannabis sativa L.	Cannabaceae	*P. Sharma 03*(FOF)	Ganja	Shrub	leaves	Juice	wild	Swollen stomach in both human and animal, Diarrhea, Gastritis	Kafle & Dani, 2022; Miya et al., 2021
Cantharellus odoratus (Schwein.)Fr.	Cantharellaceae		Champe chau/Hardayana chhati	Herb	whole plant	Cooked	wild	As vegetable	
Capsicum frutescens L.,	Solanaceae		Khursani	Herb	Fruit	Raw	Cultivated	Used as spice, Gastritis	
Carica papaya L.	Caricaceae		Mewa	Shrub	Fruit, latex	Juice	Cultivated	Edible fruit, latex is used in Identification of snake bite	Ambu et al., 2020

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Casia fistula L.	Fabaceae		Banarshot	Tree	Fruit	Juice	wild	Diarrhea, Vomiting, Constipation, Used in banana ripening, Urine retension	Kafle & Dani, 2022; Miya et al., 2021; Rijal, 2011; Sharma et al., 2014
Centella asiatica (L.) Urb.	Apiaceae	*P. sharma 04* (FOF)	Ghodtapre	Herb	Root, Leaves	Juice	wild	Urinary trouble, Tonsillitis, Fever Burn,	Acharya & Pokhrel, 2006; Bhattarai et al., 2009; Kafle & Dani, 2022; Luitel et al., 2014; Mueller-Boeker, 1993; Sharma et al., 2014; Shrestha & Dhillion, 2003; Tamang & Sedai, 2016
Chromolaena adorata (L.) R. King & H. Rob.	Asteraceae		Banmara	Herb	Leaves	Paste	wild	Cuts and Wounds	Kafle & Dani, 2022
Cissus quadrangularis L.	Vitaceae		Hadjora	Climber	Roots	Paste	Cultivated	Bone fracture of animal,	Rajbanshi & Thapa, 2019
Citrus aurantiifolia (L.) Osbeck	Rutaceae		Kagati	Shrub	Fruit	Juice	Cultivated	Kidney stone, Energy boost, Diarrhea, Edible fruits	Rajbanshi & Thapa, 2019
Clerodendrum indicum (L.) Kuntze	Lamiaceae		Agiyathi	Shrub	Root	paste	wild	Wounds, Allergy	
Clerodendrum infortunatum(L.) Kuntze	Lamiaceae		Bhatik patta	Herb	Root	Juice	wild	Tonsillitis, Fever, Chest pain	Miya et al., 2021; Mueller-Boeker, 1993
Cocos nucifera L.	Arecaceae		Nariol	Tree	Fruit	Juice	Cultivated	Hair tonic, Edible fruit, Used in marriage ceremony	Kafle & Dani, 2022
Celosia argentea var. cristata (L.)	Amaranthaceae		Surwale sag	Herb	leaves	Decoction	Cultivated	Fever, Spinach	
Curcuma caesia Roxb.	Zingiberaceae		Kalo Besar	Herb	Root	Juice	Cultivated	Swollen stomach, Tonsillitis, Back pain	Thapa, 2012
Cuscuta reflexa Roxb.	Convolvulaceae		Amarlatti/sikari lahara	Climber	whole plant	Decoction	wild	Jaundice, Piles, Fractures	Ambu et al., 2020; Ghimire, 2016; Kafle & Dani, 2022; Miya et al., 2021; Rajbanshi & Thapa, 2019; Rokaya et al., 2010; Singh, 2020; Singh et al., 2012
Cyperus rotundus L.	Cyperaceae	*P. Sharma 05* (FOF)	Motha	Herb	Flower	Massage	Roadside	Headache,	Kafle & Dani, 2022
Dalbergia sissoo Roxb. ex DC.	Fabaceae		Sissau	Tree	Latex		wild	Diarrhea, Dysentery, Used in marriage ceremony	Acharya & Pokhrel, 2006 ; Miya et al., 2021; Singh et al., 2012
Datura metel L.	Solanaceae	*P. Sharma 06* (FOF)	Dhaturo	Herb	Fruit, Leaves	Poultice, Massage	Cultivated	Asthma, Swollen leg, Swollen stomach, Dog bite,	Singh et al., 2012
Elsholtzia fruticosa (D.Don) Rehder	Lythraceae	*P. Sharma 07* (FOF)	Dhursi	Shrub	Leaves	Juice	wild	Banana ripening, Religious value	

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Euphorbia royleana Boiss.	Euphorbiaceae		Paspatko patta	Herb	leaves	Poultice	Cultivated	Chest pain in children, whooping cough	Ghimire, 2016; Miya et al., 2021
Falconeria insignis Royle	Euphorbaceae		Khirro	Tree	Bark	paste	Cultivated	Milk production,	
Ficus religiosa L.	Moraceae		Peepal	Tree	Root		wild	Religious value	
Hellenia speciosa (J.Koenig) S.R.Dutta	Costaceae		Betlauri	Herb	Root	Juice	Cultivated	Weakness, Jaundice	
Ichnocarpus frutescens (L.) W.T.Aiton	Apocynaceae		Dudhiya lathhi	Climber	Root	Powder	wild	Milk production	Kafle & Dani, 2022
Jatropha curcas L.	Euphorbiaceae		Bagandi/ Sajiwan	Tree	Fruit	Cooked	Cultivated	Lower pressure, used as vegetables	
Juniperus L.	Cupressaceae		Dhupi	Tree	Leaves, Fruit		Cultivated	Dried leaves are used in making incense, Used in marriage ceremony	
Justicia adhatoda L.	Acanthaceae	*P. Sharma 08* (FOF)	Aasuro	shrub	Leaves	Infusion, Decoction	wild	Leaves are used as fertilizer, Jaundice, cough and cold, Fever	Bhattarai et al., 2009; Luitel et al., 2014; Singh et al., 2012; Tamang & Sedai, 2016
Lablab purpureus (L.) Sweet	Fabaceae		Chhimi	Climber	Leaves	paste	Cultivated	Allergy, used as vegetables	Tamang & Sedai, 2016
Lawsonia inermis L.	Lythraceae		Mehendi	Herb	Leaves, stem	paste	Cultivated	Hair colour, Trench foot, Fungal infection	Kafle & Dani, 2022; Singh, 2020
Leucas aspera (Willd.) Link	Lamiaceae	*P. Sharma 09* (FOF)	Danduke	Herb	Flower, leaves	paste, Decoction	Agricultural land	Tonic, Tonsillitis, Fever, Skin allergy	
Leucas cephalotes (Roth) Spreng.	Lamiaceae		Dhulphi	Herb	Root	Decoction	Agricultural land	Fever	Singh, 2017
Lygodium palmatum (Bernh.) Sw.	Lygodiaceae	*P. Sharma 10* (FOF)		Climber	Leaves	Massage	Agricultural land	Headache	
Macrotyloma uniflorum (Lam.) Verdc.	Fabaceae		Gahat	Herb	Fruit	Cooked	Cultivated	Kidney stone, Cough, Agricultural food	Kafle & Dani, 2022
Mallotus philippensis (Lam.) Müll.Arg.	Euphorbiaceae		Sindure	Tree	Bark	Paste	Wild	Dysentery	
Mangifera indica L.	Anacardiaceae		Aap	Tree	Bark, Leaves, fruit	Infusion	Cultivated	Jaundice, Edible fruit, Agricultural tool, Perform Mango marriage	
Melastoma malabathricum L.	Melastomataceae	*P. Sharma 11* (FOF)	Aageri	Shrub	Fruit	After ripening	Wild	Wild edible Fruit, Typhoid, Fever,	
Melia azedarach L.	Meliaceae		Bakaino	Tree	Leaves, Bark	Juice	Cultivated	Foot and mouth disease in chicken, lower pressure	Malla & Chhetri, 2009
Mentha spicata L.	Lamiaceae		Pudina	Herb	Leaves	Juice	Cultivated	Indigestion, Gastritis, Leaves as condiment	Ghimire, 2016; Malla & Chhetri, 2009; Rokaya et al; 2010

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Mimosa pudica L.	Fabaceae		Lajoni	Herb	Root, young shoot	paste	Roadside	Wounds, Control bleeding, Tonsillitis, Diabetes	Ghimire, 2016
Mucuna pruriens (L.) DC.	Fabaceae		Kauso	Climber	Root	Juice	Cultivated	Abortion, Tonsillitis	
Musa balbisiana colla	Musaceae		Ban kera	Shrub	Fruit	Raw	wild	Fish bone/Bone pricking, Asthma	Kafle & Dani, 2022
Musa×paradisiaca L.	Musaceae		Kera	Shrub	Fruit	Raw, Cooked	Cultivated	Ripen fruits are edible, cooked like vegetables, Diarrhea, Used in ritual	Luitel et al., 2014; Malla & Chhetri, 2009; Singh et al., 2012
Nerium oleander L.	Apocynaceae	*P. Sharma 12* (FOF)	Hardyana phula	Shrub	Latex	Poultice	Agricultural land	Wounds	
Nicotiana tabacum L.	Solanaceae		Surti	Herb	Leaves	Topical	Agricultural land	Remove leech	Kafle & Dani, 2022; Miya et al., 2021; Rai, 2004
Nyctanthes arbor-tristis L.	Oleaceae		Parijat	Shrub	Leaves	Decoction	Roadside	Cold and Cough, Fever	Ambu et al., 2020; Kafle & Dani, 2022
Ocimum basilicum L.	Lamiaceae		Babari	Herb	Flower, Fruit, Laves	Decoction	Cultivated	Typhoid,	Poudel & Singh, 2016
Ocimum tenuiflorum L.	Lamiaceae		Tulasi	Herb	Leaves	Decoction	Cultivated	Cough, Cold, Fever, Gastritis, Used in the worship of Bishnu	Ambu et al., 2020; Bhattarai et al., 2009; Kafle & Dani, 2022; Singh et al., 2012
Ophioglossum reticulatum L.	Ophioglossaceae		Jibiya sag	Herb	whole plant	Juice	Wild	Diarrhea/ Dysentery, Used as vegetable	Kafle & Dani, 2022; Rajesh et al., 2013
*Opuntia monacantha*Haw.	Cactaceae		Bagiya kanda	Shrub	Latex		Cultivated	Eye infection in animal, Food aversion	
Orchid sp.(Epiphytic)	Orchidaceae		Sunakhari	Herb	Root /Stem	Poultice	Wild	Bone Breakage	Kafle & Dani, 2022
Oroxylum indicum (L.) Kurz	Bignoniaceae		Totela	Tree	Flower, Fruit Bark	Juice	Cultivated	Used as vegetables, Jaundice, Gastritis, Used in Ritual	Ghimire, 2016; Tamang & Sedai, 2016
Oryza sativa L.	Poaceae		Dhan	Herb	Rhizome	Paste, Poultice	Cultivated	Scorpion bite	
Oxalis corniculata L.	Oxalidaceae		aamti	Herb	whole plant	Juice	Agricultural land	Stomach disorder, Used as Spinach	Ghimire, 2016; Malla et al., 2015; Rokaya et al., 2010
Paederia foetida L.	Rubiaceae		Gulatti	Climber	Root	Juice	Wild	Tonsillitis , Appetizer	Kafle & Dani, 2022
Perilla frutescens (L.) Britton	Lamiaceae		Silam	Herb	Seed	Raw	Agricultural land	Used as spice	Ambu et al., 2020
Phoenix acaulis Roxb.	Arecaceae		Thakkal	Shrub	Fruits	after ripening	Wild	Used in making traditional drink	
Phyllanthus emblica L.	Phyllanthaceae		Rhikhiya	Tree	Fruits, Leaves/Stem	Raw	Wild	Gastritis, Edible fruits, Used in Ritual	Acharya & Acharya, 2009; Bhattarai et al., 2009; Luitel et al., 2014;
Piper longum L.	Piperaceae	*P. Sharma 13* (FOF)	pipila	Climber	Root, Fruit	Juice	Agricultural land	Cough, Fever, Headache	Malla et al., 2015; Miya et al., 2021; Shrestha et al., 2016

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Plumbago zeylanica L.	Plumbaginaceae		Chitu	Herb	Leaves, Bark	Juice	Cultivated/ Wild	Wounds, Removing scars, Swollen stomach, Food aversion	Ambu et al., 2020; Bhattarai et al., 2009
Pogostemon benghalensis (Burm.f.) Kuntze	Lamiaceae		Utjara	Shrub	Leaves, Root	Juice	Agricultural land	Treatment of lice in Goat, Fever	Bhattarai et al., 2009; Dangol & Gurung, 1991; Kafle & Dani, 2022; Malla et al., 2015
Premna serratifolia L.	Lamiaceae		Gineri	Shrub	Bark , Latex	Decoction	Agricultural land	Burning sensation ,Dysentery	
Psidium guajava L.	Myrtaceae		Biloki	Tree	Leaves, Bark	Decoction	Cultivated	Diarrhea, Dysentery	Bhattarai et al., 2009; Luitel et al., 2014; Malla & Chhetri, 2009; Singh, 2020; Tamang, 2003; Tamang & Sedai, 2016
Ricinus communis L.	Euphorbiaceae	*P.Sharma 14* (FOF)	Andi	Shrub	Root , Latex, seed	Juice , Topical	Roadside	Amoebic dysentery , Hand or foot cracks	Bhattarai et al., 2009; Kafle & Dani, 2022; Singh et al., 2012
Saccharum officinarum L.	Poaceae		Ukhu	Herb	Stem	Juice	Cultivated	Edible Stem, Jaundice, Used in Marriage ceremony	Ghimire, 2016; Malla & Chhetri, 2009; Miya et al., 2021; Poudel & Singh, 2016; Singh, 2020
Scoparia dulcis L.	Plantaginaceae	*P. Sharma 15* (FOF)	Balyari	Herb	Stem/Root	Juice	Wild	Cough, Diarrhea	Kafle & Dani, 2022
Semecarpus anacardium L.f.	Anacardiaceae		Bhala	Tree	Fruit	Topical	Wild	Skin irritation due to latex	Kafle & Dani, 2022
Senegalia catechu (L.f.) P.J.H. Hurter & Mabb.	Fabaceae		Khair	Tree	Core stem	Decoction	Wild	Cough, Used in colouring the Traditional drink, Bone fracture	Ambu et al., 2020; Kafle & Dani, 2022
Senna tora (L.) Roxb.	Fabaceae	*P. Sharma 16* (FOF)	chekor	Herb	Leaves, Seed	Juice	Roadside	Kidney stone, Used as vegetables,	Rajesh et al., 2013
Sesamum indicum L.	Pedaliaceae		Til	Herb	Seed	Topical	Agricultural land	Used as spice, Reduce pain, Ringworms, Used in ritual	Rai, 2004
Shorea robusta C.F.Gaertn.	Dipterocarpaceae		Sakhuwa	Tree	Bark, Leaves, Latex	Smoke	Wild	Used in making incense, Leaves are used in making plate, Used in Ritual	Ghimire, 2016; Miya et al., 2021; Rijal, 2011; Sharma et al., 2014
Sida cordifolia L.	Malvaceae	*P. Sharma 17* (FOF)	Seto balu	Herb	Root	Raw	Agricultural land	Boils, Toothache	Miya et al., 2021; Rajbanshi & Thapa, 2019
Sida rhombifolia L.	Malvaceae		Kalo balu	Herb	Root	Infusion	Agricultural land	Stomache, Wounds	Miya et al., 2021 ; Mueller-Boeker, 1993; Rijal, 2011
Smilax aspera L.	Smilacaceae	*P. Sharma 18*(FOF)	Kukurdaino	Climber	Stem	Paste	Wild	Scorpion bite	
Solanum torvum Sw.	Solanaceae		Badmi	Shrub	Fruits, Leaves	Decoction	Agricultural land	Fever, Cough, Antidote against poisoning	
Spondias pinnata (L.f.) Kurz.	Anacardiaceae		Amaro	Tree	Burned fruit pulp		Wild	Common cold	Kafle & Dani, 2022
Syzygium aromaticum (L.) Merr.&L.M.Perry	Myrtaceae		Nong	Tree	Fruit	Put in Teeth	Wild	Toothache, Used as Spice	Ghimire, 2016; Miya et al., 2021
Syzygium cumini (L.)	Myrtaceae		Phadim/Jamuna	Tree	Bark of Fruits	Juice	Cultivated	Diarrhea, Diabetes , Edible	Ambu et al., 2020; Bhattarai et al.,

Scientific name	Family	Voucher number	Local name (Danuwari)	Life form	Parts used	Form of use	Habit	Indigenous uses	Similar use reports
Skeels								fruits	2009; Kafle & Dani, 2022; Miya et al., 2021; Rijal, 2011; Singh, 2020; Singh et al., 2012
Syzygium nervosum DC.	Myrtaceae		Thumki jam	Tree	Leaves	Fumigation	Wild	Sinusitis	Kafle & Dani, 2022
Tagetes erecta L.	Asteraceae		Saipatri	Herb	Flower	Decoction	Cultivated	Tonsillitis, Used in Festival	Ambu et al., 2020
Terminalia alata B.Heyne ex Roth	Combretaceae		Saj	Tree	Bark	Infusion	Wild	Dysentery	
Terminalia bellirica (Gaertn.) Roxb.	Combretaceae		Barro	Tree	Fruits	Juice	Wild	Used in making Traditional drink, Gastritis	Ambu et al., 2020; Poudel & Singh, 2016
Terminalia chebula Retz.	Combretaceae		Harro	Tree	Bark of Fruits	Powder , Infusion	Wild	Cough, Fever, Tonsillitis, Gastritis, Tonic	Acharya & Acharya, 2009; Kafle & Dani, 2022; Poudel & Singh, 2016; Rajbanshi & Thapa, 2019; Singh et al., 2012
Tinospora sinensis (Lour.) Merr.	Menispermaceae		Guuji	Climber	Root ,Stem	Decoction, juice	Wild	Stomach disorder	Singh, 2020
Trichosanthes cucumerina L.	Cucurbitaceae		Ban ghiraula	Climber	Fruit, Seed	Powder	Wild	Jaundice, Yellowish teeth, Sinusitis, Headache	Kafle & Dani, 2022
Urtica dioica L.	Urticaceae	*P. Sharma 19*(FOF)	Sisno	Shrub	Whole plant	Decoction , Cooked	Wild	Lower pressure, Diarrhea, Used as vegetables, Rheumatism	Kafle & Dani, 2022; Miya et al., 2021; Poudel & Singh, 2016; Shah & Laminachhane, 2017; Shrestha et al., 2016; Thapa, 2012
Vitex negundo L.	Lamiacae		Simali	Shrub	Leaves	Massage	Agricultural land	Headache, Used in marriage ritual	Kafle & Dani, 2022
Zingiber cassumunar Roxb.	Zingiberaceae		Phachang	Herb	Rhizome	Infusion	Cultivated	Jaundice, Swollen stomach in both Human and livestock	
Ziziphus mauritiana Lam.	Rhamnaceae		Bayer	Tree	Root	Juice	Wild	Typhoid	Kafle & Dani, 2022; Malla et al., 2015; Pangeni et al., 2020

Journal of Plant Resources (2025) **23**(1), 107-130
https://doi.org/10.3126/bdpr.v23i1.83358

Review Article

Invasive Alien Plant species (IAPs) for Sustainable Pest Management: A Review

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Abstract

Invasive alien plants (IAPs) threaten biodiversity and agricultural productivity via disruption of ecological processes. Managing IAPs is challenging as they spread across Nepal. Prevention is now challenging, and eradication is only feasible in small, managed areas. IAPs have great potential as sustainable source of bio-pesticides. The use of synthetic pesticides for controlling agricultural pests, household insects, and mosquitoes has significant drawbacks. They are expensive, persist long-term in soil and food, negatively impact human health and environment, and lead to pest resistance. Hence, IAPs can be the alternative for synthetic pesticides. This review was conducted to examine the insecticidal activities of Nepal's IAPs to generate alternative solutions for pest and IAPs management by utilizing them. The study involved a comprehensive literature review of the insecticidal properties of 30 IAPs of Nepal. Data were collected from various published studies and the focus was on identifying the plant parts used for formulations, the insect and their stages targeted, and the overall effectiveness of different extract formulations. Among the 30 IAPs of Nepal, 18 species have been studied at many countries for their insecticidal properties against various insects, including mosquitoes and agricultural pests. Most studies were conducted in India and Nigeria. Leaves were the most frequently used plant part for formulations. The larval stage of insects was the preferred stage for studying insecticidal properties. Essential oils were found to be more effective than other extract formulations. All studies demonstrated toxicity to different insects at different level. IAPs of Nepal possess insecticidal activity against harmful insects and pests, offering potential for sustainable pest management.

Keywords: Bio-pesticides, Insecticides, Invasive plants, Mosquito

Introduction

Synthetic pesticides pose significant issues related to human health, soil quality and impact on non-target organisms thereby causing devastating impact on floral, faunal and microbial diversity. Their persistent nature, high cost, environmental impact, and development of pest resistance have raised concerns, highlighting the need for eco-friendly, cost-effective, and sustainable bio-pesticides with specific modes of action (Hicks et al., 2018). Unlike synthetic insecticides, which have eco-toxicological impacts, negatively affect non-target organisms, and cause genetic drift in targeted species; bio-pesticides offer a safer alternative (Ayilara et al., 2023) They can repel insect pests, disrupt their development, affect reproduction, or kill live organisms on contact. Bio-pesticides are eco-friendly and accessible to resource-poor smallholder farmers and locals in regions with food insecurity (Kumar et al., 2021). Bio-pesticides exhibit various modes of action, such as denaturing protein structures, inhibiting growth rates, causing metabolic disorders, and releasing bioactive compounds (Sparks & Nauen, 2015).

Several plants and micro-organisms have been utilized for bio-insecticide formulations. For example, *Azadirachta indica* A. Juss. (neem) is used by farmers in developing countries as an insecticide. It contains the bioactive compound azadirchtin, which exhibits pronounced insecticidal activity, affecting egg hatchability, fecundity inhibition, oviposition repellence, larval, pupal, and adult lifespan, insect growth regulation, ovicidal effects, and anti-feedant properties (Chaudhary et al., 2017). Extracts from various plants (Ŗengül Demirak &

Canpolat, 2022), such as *Adhatoda vasica* L. leaves, *Annona squamosa* L. seeds, *Nerium oleander* L. stem, *Volkameria inermis* L., *Pongamia pinnata* (L.) Pierre seeds, *Prosopis chilensis* (Molina) Stuntz stems, *Vitex negundo* L. leaves, *Madhuka indica* J.F. Gmel, *Azadirchta indica* and *Aegle marmelos* (L.) Corrêa had shown significant insecticidal activity against *Nilaparvata lugens* (Stal) (Hiremath et al., 1997). Additionally, ethanol extracts from tropical plants, including *Nikotina tobaccum* L., *Dioscorea pallens* Schltdl., *Cuscuta americana* L., *Bontia daphnoides* L., *Capsicum annuu* L., *Artocarpus altilis* (Parkinson) Fosberg, *Cyclopeltis semicordata* (Sw.) J.Sm., *Hibiscus rosa -sinensis* L., *Annona reticulate* L., *Mimosa pudica* L., *Gliricidia sepium* (Jacq.) Kunth and *Chromolaena odorata* (L.) R.M.King & H.Rob. have demonstrated insecticidal activity with 50-100% mortality of *Tribolium confusum* Jacquelin du Val, 1863 adults (Williams & Mansingh,1993).

Building upon the use of varied plants species for the bio-pesticides formulations, IAPs offer twin benefit by addressing the need of sustainable pest management and the ecological burden of IAPs themselves. IAPs is emerging as a unavoidable environmental problem, threatening biodiversity and agricultural productivity (Roy et al., 2024). In Nepal, 182 alien plant species have naturalized, with 30 classified as invasive (Shrestha et al., 2024).Their distribution and impact is increasing, complicating the management of these species (Shrestha & Shrestha, 2021). Among these, four species - *Lantana camara* L., *Mikania micrantha* Kunth, *Pontederia crassipes* Mart. and *Chromolena odorata* (Spreng.) R.M.King & H.Rob- are listed among the world's 100 invasive species (Shrestha, 2016). Also, recently reported *Leucaena leucocephala* (Lam) de Wit is one of the 100 worst invasive species (Shrestha et al., 2024). The invasive species problem is global but is particularly pronounced in developing countries like Nepal, where management efforts are hampered by a lack of expertise and limited resources.

Invasive plant species produce allelopathic compounds (Kalisz et al., 2021) which have potential to suppress native plants. Also, invasive plants has high chemical diversity and novel secondary metabolites than native plants (Macel et al., 2014). Wide variety of phenolic compounds, alkaloids, volatile organic compounds from the invasive plants have shown the bio-medical importance presenting anti-cancer, anti-bacterial, anti-fungal, anti-oxidant activities. These secondary metabolites can also have the potential source of bio-pesticides. The review aims to explore the management of problematic IAPs in Nepal through their economic utilization for insecticide formulation. Authors synthesized existing but scattered literature on the insecticidal activity of various parts of IAPs, specifically those invasive in Nepal. The paper discusses the prospects and opportunities for using IAPs as bio-insecticides for controlling agricultural pests, protecting the environment, and addressing human health concerns. Finally, the paper highlights trends in bio-pesticide use, identify research gaps, and recommend bioassay-guided studies of IAPs as insecticide for future research.

Materials and Methods

Search strategy

The information presented in this review was sourced from journal articles relevant to the insecticidal properties of IAPs in Nepal. Scientific papers were obtained from diverse sources, including Google Scholar, PubMed, DOAJ and EBSCO. Systematic keywords used in the search included "invasive alien plants," "insecticidal," "mosquitocidal potential," along with the scientific names and synonyms of each plant reported to possess insecticidal properties. Boolean operators (OR) were used to refine the search for more focused and productive results. The literature search was conducted during May 2024.

Inclusion and exclusion criteria

Only literature focusing on insecticidal properties was included. Out of 143 published papers retrieved, 60 were reviewed. Excluded papers were those published before 2014, those involving plants not declared invasive in Nepal, ethno-botanical studies, clinical trials, studies on non-IAPs of Nepal, insect repellency, ovicidal effect; and those focused on impact and distribution.

Data extraction and analysis

Data recorded included the percentage of insect mortality, LC_{50} and LC_{90} values (converted to ppm where possible), country of study, IAP names, plant parts used, formulations, target insects, developmental stages at which the formulation was applied, highest percentage mortality, observation periods, and reported LC_{50} and LC_{90} values. Microsoft tools and reference manager (Zotero) were used for data analysis and reference management, respectively. The ranking of studied articles was assessed using the SCImago ranking website. But both ranked and non-ranked journal were studied.

Results and Discussion

Case studies of insecticidal potential of IAPs in different countries

From the literature survey, 60 studies published in the last ten years globally, reporting on the insecticidal activities of 18 plant species that are invasive to Nepal. The highest numbers of published case studies were from India and Nigeria, with 20 and 15 studies, respectively. Other countries, such as Egypt, Brazil, China, Ethiopia, Ghana, Kenya, Colombia, Thailand, Vietnam, Ivory Coast, Burkina Faso, Slovenia, the Philippines, and the United States, each contributed fewer than five studies. No records were obtained from Nepal regarding the insecticidal potential of IAPs (Figure1), despite extensive studies on their distribution and impact (Pandey et al., 2020). It can be hypothesized that the large number of research papers from India, Nigeria, and other developing countries is due to the availability of those IAPs in these countries than other; and the need of local people to manage insect pests of agricultural, environmental and medical importance.

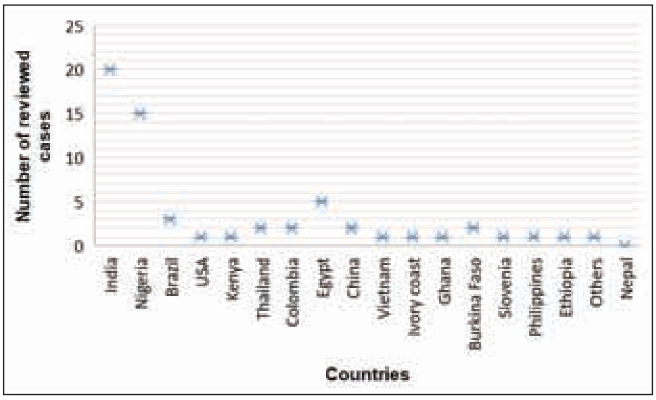

Figure 1: Case studies of insecticidal potential of IAPs in different countries

Insecticidal potential of different IAPs of Nepal

The published articles (Table 1) on the insecticidal potential of Nepal's IAPs, represents species names, plant parts used, test sample formulations, targeted insects and stages, percentage mortality, and/or LC_{50}, LC_{90} and duration of observation. Information regarding the phytochemicals data and methods of nanotechnology-based formulations are not mentioned here, but presented the dependency of insecticidal effect on concentration and formulation of test sample, biological assay and post-exposure time for varied insects. Additionally, only a few cases were recorded to have performed toxicity tests on albino mice or brine shrimp, which are also excluded from the data. Gas Chromatography Mass Spectrometry (GC-MS) data was not the focus point of this review.

These studies utilized different plant parts to formulate essential oils, crude drugs, or extracts with various solvents. Notably, no studies were found that employed bioassay-guided fractionation or examined effects on non-target organisms. This limits the identification of potential compound responsible for the insecticidal activities and maintenance of regulatory standards and environmental integrity. Most studies used sample sizes of 20-25 for mosquito species and 10 for others. The World Health Organization (WHO) methods were commonly used for mosquito applications, with slight modifications based on available instruments. Other insects, both adults and larvae, were studied for contact toxicity, fumigant toxicity, anti-feedant activity, and habitat alteration. The lethal concentration in all cases was reported with a 95% confidence level for significant differences.

Table 1: Insecticidal potential of different invasive alien plant species of Nepal

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
1	*Lantana camara* L. *(Verbenaceae)*	Leaves	Methanol (sachet)	Mosquito species	Larvicidal (2,3 Instar)	40 ± 1.91 at 1 mg/ml	NM	NM	48 h	(Kandalkar et al., 2020)
		Leaves	Hydro-distillation	*Anopheles subpictus* Grassi *Aedes aegypti* L. *Culex quinquefasciatus* Say	Larvicidal (4 Instar)	94-96% at 100 ppm	45.32 ± 1.66 ppm	96.62 ± 1.99 ppm	24 h	(Sharma et al., 2021)
		Leaves Stem Flower Synergy	Methanol (cold maceration)	*Aedes aegypti* L.	Larvicidal (4instar)	100% at 250, 500 and 1000 ppm	22.55 ppm 1695.51 ppm No activity >2000	96.44 ppm 9643.95 ppm No activity >2000	24 h	(Eze et al., 2022)
		Leaves	Methanol Aqueous (maceration)	*Anopheles* sp.	Larvicidal	100% at 100 mg/ml (Aq)	22.91 ppm 17.38 ppm	NM	6 h	(Ekemezie,2021)
		Leaves Stem	Hexane Ethanol Hexane Ethanol	*Aedes aegypti* L.	Larvicidal (4 instar)	100% at 1000 ppm	118.5 ppm - 89.62 ppm -	159.96 ppm - 125.89 ppm -	24 h	(Sharma et al.,2016)
		Leaves	Chloroform Petroleum ether Methanol (soxhlet)	*Aedes aegypti* L. *Culex quinquefasciatus*Say. *Aedes aegypti* L. *Cu. Quinquefasciatus* Say *Aedes aegypti* L. *Culex Quinquefasciatus* Say	Larvicidal (4th or late 3rd instar)		>300 ppm 55.92 ppm 74.93 ppm 10.63 ppm >300 ppm -	>300 ppm 146.61 ppm 149.4 ppm 92.98 ppm >300 ppm -	48 h	(Hari &Mathew,2018)
		Leaves	Essential oil loaded nanoemulsion	*Aedes aegypti* L.	Larvicidal I-instar II-instar III-instar iv- instar Pupicidal Adulticidal	-	 18.18 ppm 23.34 ppm 29.73 ppm 38.94 ppm 45.30 ppm LD50: 11.95 mg/cm^2	 46.96 ppm 56.48 ppm 71.67 ppm 82.02 ppm 85.99 ppm LD90: 47.72 mg/cm^2	-	(Udappusamy et al.,2022)
		NM	 Acetone Ethanol Hexane	*Anopheles gambie* Giles Sensitive strain local strain	Larvicidal Sensitive Local Sensitive Local Sensitive Local	100% in 160-320 ppm of hexane extract.	 32.62 ppm 106.09 ppm 29.23 ppm 122.82 ppm 15.94 ppm 20.19 ppm	 180.29 ppm 91.19 ppm 63.13 ppm 202.34 ppm 42.65 ppm 49.29 ppm	24 h	(Wangrawa et al.,2016)
		Leaves	Methanol	*Anopheles arabiensis* Patton	Larvicidal (III &IV) Adulticidal (3-5day, Nonblood fed)		89.48 ppm 0.42 % mg/l	178.02 ppm 0.81 % mg/l	24 h	(Wangrawa et al.,2023)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
			Acetone		Larvicidal Adulticidal		58.72 ppm 0.43 % mg/l	143.97 ppm 0.80 % mg/l		
		Different parts	Hydrodistillation	*Aedes aegypti* L.	Larvicidal RS DS		 49 ppm 41.7 ppm	 - -	24 h	(Luz et al.,2022)
		NM	Aqueous extract	Pest of cabbage: *Plutella xylostella* L. *Brevicoryne brassicae* L. *Hellula undalis* Fab Araneae Coccinellidae	Adulticidal	Less densities were observed in untreated plot when compared with the treated plot. All the plants help in pest suppression.			4 weeks	(Amoabeng et al.,2020)
		Fresh leaves	Hydro-distillation	Rice weevil: *Sitophilus oryzae* L.	Adulticidal	-	9.81 mg/cm^2	41.20 mg/cm^2	72 h	(Tawfeek et al.,2021)
		Leaves	Hexane, Ethyl acetate, Acetone Methanol (as all are dissolved before application.)	Stored grain pest: *Sitophilus oryzae (L.),* *Callosobruchus chinensis (Fab.),* *Tribolium castaneum (Herbst.)*	Adulticidal Fumigant assay/ contact toxicity assay	100% mortality was observed in *C. chinensis* after 7 days.	128.07 ppm 0.16 mg/cm^2 130.32 ppm 0.14 mg/cm^2 178.7 ppm 0.21 mg/cm2	- - - - - -	24 h	(Rajashekar et al.,2014)
		Leaves	Essential oil	*Pomacea canaliculata* Lamark *Gyraulus convexiusculus* T. Hutton *Tarebia granifera* Lamark *Aedes aegypti* L. *Aedesalbopictus*Skuse *Culex quinquefasciatus*	Adulticidal (Mollusca) ,, ,, Larvicidal ,, ,,		23.6–40.2 ppm 7.9–29.6 ppm 15.0–29.6 ppm 15.1–29.0 ppm, 26.4–53.8 ppm 20.8–59.3 ppm	- - - 29.27-62.93 ppm 46.35-81.83 ppm 64.43-88.70 ppm	24 h	(Huy Hung et al.,2021)
		Leaves	Hydrodistillation	*Tribolium castaneum* Herbst *Lasioderma serricorne* Fabricius *Callosobruchus chinensis* L.	Adulticidal Fumigant/ Contact toxicity		16.70 ppm 8.93 mg/cm2 4.141 ppm 4.82 mg/cm2	23.21 ppm 13.54 mg/cm2 10.91 ppm 17.47 mg/cm2 27.36 ppm	24 h	(Aisha et al.,2024)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
							6.25 ppm 6.24 mg/cm2	6.27 mg/cm2		
		Leaves	Juice	*Aedes aegypti* L.	Larvicidal (III &IV)	-	52.06 ppm	104.33 ppm	24 h	(Sharma et al.,2023)
				Anopheles subpictus Grassi			70.91 ppm	218.10 ppm		
				*Culex quinquefasciatus*Say			47.47 ppm	106.70 ppm		
		Leaves	Copper nanoparticles	*Anopheles multicolor* Combouli	Larvicidal (III instar)	100% at 20 ppm	12.5 ppm	18.4 ppm	24 h	(Abd El Hafiz Hassanain et al..2019)
			Petroleum ether extract			140 ppm	63.5 ppm	119.9 ppm		
2.	*Parthenium hysterophorus* L. (Asteraceae)	Leaves	Methanol sachet	Mosquitospecies	Larvicidal (III&IV instar)	78 ± 1.70% at 1mg/ml	NM	NM	48 h	(Kandalkar et al.,2020)
		Leaves	Hexane, Acetone, Petrolium ether Ethanol	*Anopheles arabiensis* Patton	Larvicidal (IV instar)	Stem Petroleum ether: 90% at 360 ppm; Hexane: 85-97% at 480 ppm. (Higher) <50% in ethanol	368 ppm 478 pmm 393.1 ppm	809.2 ppm 1358.2 ppm 1352.6 ppm	24 h	(Tarekegn et al.,2021)
		Stem					112.4 ppm 259.4 ppm 701.1 ppm	331.9 ppm 1814.6 ppm 701.1 ppm		
		Root					87.9 ppm 342.1 ppm 10.7 ppm	338.3 ppm 769.7 ppm 105.5 ppm		
		Leaves	Methanol	*Aedes aegypti* L.	Larvicidal (3^{rd} and 4^{th} instar)	66 ± 0.44% at 300 ppm	-	-	72 h	(Jayaraman et al.,2023)
		Whole plant	Parthenin: Ethylene glycol Azide	*Anopheles gambiae*	Larvicidal (III instar)		154 ppm 37 ppm 66 ppm	313 ppm 698 ppm 282 ppm	72 h	(Milugo et al.,2021)
3.	*Chromolena odorata* (Spreng.) R.M.King & H.Rob (Asteraceae)	Leaves	Petrolium ether, Chloroform Ethanol (maceration)	*Culex quinquefasciatus*	Larvicidal (IV instar)	90% in 2.5 and 5 ppm petroleum ether.	0.16 ppm 0.24 ppm 0.64 ppm	9.19 ppm 52.30 ppm 38.75 ppm.	72 h	(Kumar et al.,2023)
		Leaves	Methanol (maceration)	*Anopheles stephensi*	Larvicidal (III and IV instar)	100% at 1000 ppm	1613 ppm	8306 ppm	24 h	(Sukhthankar et al.,2014)
				Culex quinquefasciatus		220 ppm	43 ppm	110 ppm		
				Aedes aegypti L.		900 ppm	138 ppm	463 ppm		
		Leaves	Methanol (maceration)	*AnophelesGambiae*	Larvicidal (III &IV) Pupicidal adulticidal	100 % at 160 mg/l to larva and pupa but not adult	31.33 ppm 53.61 ppm 296.20 ppm	95.73 ppm 117.66 ppm 2221.05 ppm	24 h	(Ileke &Olabimi,2019)
		Leaves Stem Root	Aqueous	*Culex pipiens*	Adulticidal	Whole part most potent	-	-	Spray tech.	(Sasan et al.,2021)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
		Leaves	Fresh juice	*Aedes* *Culex*	Larvicidal (III instar)	100% at 4 mg/ml	-	-	72 h	(Rathy et al.,2015)
		Leaves	Ethanol (soxhlet)	*Anopheles gambiae*	Adulticidal	60% at 5 mg/ml	4.52 ppm	8.32 ppm	24 h	(Afolabi et al.,2018)
		Leaves	Methanol Acetone Water	*Spodoptera litura* Fabricius (tobacco eating)	Ovicidal Antifeedant larvicidal	73.33 ± 0.57% 82.45 ± 0.16% 68.33 ± 0.05% Less mortality "	-	-	96 h	(Gorawade et al.,2022)
		Leaves	Aqueous n-butanol	*Anopheles gambiae*	Larvicidal (III and early IV)	-	3497.27 ppm 3117.97 ppm	9707 ppm 9283.08 ppm	24 h	(Imam et al.,2018)
		Leaves	Fresh pulverized juice	*Periplanata Americana* L.	Adulticidal	36.63 % at 1 ml	-	-	48 h	(Udebuani et al.,2015)
		Leaves	Aqueous AgNO3 nanoparticles	*Culex quinquefasciatus*	Larvicidal III/IV	30.67 ± 0.33 26.67 ± 0.33 (At 250 ppm) 81.33 ± 0.88 70.67 ± 0.88	396.8 ppm (III) 448.3 ppm (IV) 95.9 ppm (III) 166.4 ppm (IV)	716.8 ppm 803.9 ppm 337.5 ppm (III) 438.7 ppm (IV)	24 h	(Elemike et al.,2017)
4.	*Spergula arvensis* L. (Caryophyllaceae)	Whole plant	Methanol Hexane Dichloromethane (Soxhlet)	*Annopheles. Culicifacies* Giles	Larvicidal (IV instar)	100% at 40 µg/ml at Methanol	6.84 ppm 2265.007 ppm 1951.79 ppm	24.816 ppm 5738.69 ppm 7238.69 ppm	24 h	(Sogan et al.,2021)
5.	*Argemone mexicana* L. (Papaveraceae)	Leaves	Methanol (maceration)	*Anopheles subpictus* Grassi	Larvicidal (IV instar)	83.33 ± 0.37% at 400 ppm	224.45 ppm	420.34 ppm	24 h	(Maheshwari et al.,2023)
6.	*Ageratum conyzoides*L. (Asteraceae)	Leaves	Ethanol (maceration)	*Culex quinquefasciantus*	Larvicidal	61.7% at 0.6 ml	-	-	48 h	(Ojianwuna et al.,2021)
		Leaves and different colored flower	Ethanol (maceration) EO (hydrodistillation)	*Aedes aegypti* L.	Larvicidal (IV instar) Adulticidal (LP)	No effect	- LD_{50}=0.84%	- LD_{90}=1.76%	24 h	(Pintong et al.,2020)
		Leaves	Petrolium ether (Soxhlet)	*Aedes aegypti* L. *Anopheles stephensi* *Culex quinquefasciatus*	Larvicidal (IV instar)	78 % 77% 73% (150 ppm)	1.92 ppm 1.95 ppm 1.97 ppm	2.99 ppm 2.33 ppm 2.37 ppm	24 h	(Ramasamy et al.,2021)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
		Aerial parts	Hydrodistillation EO	*Sitophilus oryzae* *Rhyzopertha dominica* Fabricius *Tribolium castaneum* Herbst	Adulticidal Contact toxicity/ fumigant toxicity	73.6± 1.2%/ 100% 60 20g/kg	>10000 ppm >10000 ppm >10000 ppm	>10000 ppm 10000 ppm >10000 ppm (LC_{95})	12days	(Nenaah,2014)
		Leaves	Steam distillation	*Anopheles gambiae*	Larvicidal (III) Adulticidal	100% at 0.30 mg/ml 3.3% at 0.05 mg/ml	190 ppm -	420 ppm -	48 h	(Adelaja et al.,2023)
		NM	Aqueous extract	Pest of cabbage: *Plutella xylostella* L. *Brevicoryne brassicae* L. *Hellula undalis* Fab Araneae) Coccinellidae	Adulticidal	Less densities were observed in untreated plot when compared with the treated plot. All the plants helped in pest suppression. 4 weeks			4 weeks	(Amoabeng et al.,2020)
7.	*Bidens pilosa* L. (Asteraceae)	Leaves	Ethanol (maceration)	*Culex quinquefasciantus*	Larvicidal	100% at 0.6 ml	-	-	48 h	(Ojianwuna et al.,2021)
8.	*Ageratina adenophora* (Spreng.) R.M.King & H.Rob (Asteraceae)	Leaves	Methanol Chloroform Petroleum ether	*Culex quinquefasciatus*	Larvicidal (III instar)	55% 100% 100% At 500 ppm	501.93 ppm 335.10 ppm 186.98 ppm	611.29 ppm 768.36 ppm 514.24 ppm	24 h	(Samuel et al.,2014)
		Leaves & Stems	Petroleum ether (Partitioning) Com:1 Com:2 Com:3	*Psoroptes cuniculi* Delafond	Adult (Acaricidal)	100% at 4-6 h at 50 % extract	0.35% 0.07 % 0.58 %	NM	4-6 h	(Nong et al.,2014)
9.	*Mesosphaerum suaveolens* (L.)Kuntze (Lamiaceae)	Leaves	Ethanol (soxhlet)	*Anopheles gambiae*	Larvicidal Pupicidal Adulticidal	100% at 0.8% 100% at 1% 100% at 0.8%	NM	NM	48Hrs „ 4hrs	(Obembe et al.,2024)
		Leaves	Hydrodistillation	*Anopheles* sp.	Larvicidal (III&IV) Adulticidal	100% at 50 ppm 44% dead at 5ml in 90 min.	3.58 ppm 6.20 ppm	13.83 ppm 28.28 ppm	6 h 1.5 h	(Duniya et al., 2022)
		Leaves	Ethanol Aqueous (cold maceration)	*Anopheles* sp.	Larvicidal (I-IV)	75% at 0.1 mg 0.50% at 1 mg	-	-	6 h 12 h	(Sani et al.,2021)
		Leaves	Methanol Petroleum ether	*Culex quinquefasciatus*/ *Aedes aegypti* L.	Larvicidal (III or IV)	-	>300 ppm >300 ppm 38.39 ppm 64.49 ppm	>300 ppm >300 ppm 78.11	48 h	(Hari &Mathew,2018)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
			Chloroform (soxhlet)				41.39 ppm >300 ppm	ppm 139.28 ppm 81.68 ppm >300 ppm		
		NM	Acetone Ethanol Hexane (maceration)	*Anopheles Gambiae Sensitive/ local strain*	Larvicidal ((III&IV)	100% at 320 ppm of hexane and acetone extract	95.66 ppm 201.66 ppm 78.88 ppm 206.49 ppm 77.33 ppm 109.72 ppm	196.76 ppm 341 ppm 193.49 ppm 339.97 ppm 120.7 ppm 179.32 ppm	24 h	(Wangrawa et al.,2016)
		Leaves	EO (Clevenger)	*Aedes aegypti* L.	Larvicidal (III)	100% at 100 ppm	32.85 ppm	69.27 ppm	24 h.	(Moola et al.,2023)
		Leaves	Aqueous n-butanol	*Anopheles gambiae*	Larvicidal (III&IV)	-	2613.01 ppm 2167.92 ppm	8553.77 ppm 7307.60 ppm	24 h	(Imam et al.,2018)
		Leaves	EO Steam distillation	*Anopheles gambiae*	Larvicidal (III) Adulticidal	100% at 0.30 mg/ml 36.67% at 0.50 mg/ml	240 ppm -	320 ppm -	24 h	(Adelaja et al.,2023)
		Leaves	Aqueous	*Helicoverpa armigera* Hubner	Larvicidal		30.25%	748.61%	72 h	(Bini et al.,2023)
		Leaves	EO nanoemulsion	*Culex quinquefasciatus*	Larvicidal (III)	100% at 250 ppm	102.41 ppm	168.03 ppm	24 h	(Peniche et al.,2022)
		Leaves	Acetone Methanol Hexane Aqueous	*Culex quinquefasciatus*	Larvicidal (IV)	59.58% 52.92% 90.42% 59.58% At 250 mg/ml	>10000 ppm >10000 ppm >10000 ppm >10000 ppm	>10000 ppm >10000 ppm >10000 ppm >10000 ppm	72 h	(Aremu et al.,2022)
		Leaves	EO Steam distillation	*Spodoptera frugiperda* J.E. Smith	Larvicidal	6.67 ± 0.5%	-	-	72 h	(de Menezes et al.,2020)
		Leaves	Hexane, Methanol, Acetone, Isopropanol Di-methyl-sulphoxide (soxhlet)	*Aedes albopictus*	Larvicidal (III)	-	689.69 ppm 310.47 ppm 258.39 ppm 349.77 ppm 569.73 ppm	-	24 h	(Yadav et al.,2014)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
10.	*Tithonia diversfolia* (Hemsl) A.Gray (Asteraceae)	Leaves	Methanol Aqueous (maceration)	*Annopheles* sp.	Larvicidal	100% at 100 mg/ml at 4 hrs.	>10000 ppm >10000 ppm	-	6 h	(Ekemezie,2021)
		Leaves	Butanol eluent Crude extract	*Spodoptera frugiperda*	Larvicidal (II and neonate) (Diet incorporation toxicity bioassay)	22.22% 13.33%	-	-	72 h	(Oluwamodupe et al.,2024)
		Leaves	Hexane 70% ethanol Dichloromethane Ethyl acetate Butanol Aqueous	Leaf-cutter ant *Atta cephalotes* L.	Adulticidal	- - 70-90% at 1000 ppm - - -	-	-	14 days	(Pantoja-Pulido et al.,2020)
		Stem and leaves	Green manure mulch	*Atta cephalotes*	Adult	50% decline in ant activity.	-	-	12 weeks.	(Rodríguez et al.,2015)
11.	*Pontederia Crassipes* Mart. (Pontederiaceae)	Leaves	Hexane Benzene Ethyl acetate Methanol Aqueous (soxhlet)	*Culex quinquefasciatus*	Larvicidal (IV)		80.54 ppm 1371.78 ppm 645.33 ppm 135.70 ppm 983.18 ppm	1433.89 ppm 2398.85 ppm 1744.50 ppm 777.21 ppm 2297.08 ppm	48 h	(Annie et al.,2015)
		Leaves	Ethanol (75%) Petroleum ether	*Culex pipiens* L.	Larvicidal (III)	100% at 3000 ppm 1000 ppm	-	-		(Hasaballah,2015)
		Root				2000 ppm 500 ppm				
		Whole plant	Acetone Ethanol	*Aphis craccivora* Koch	Adulticidal		64 ppm 140 ppm	-	24 h	(Abdelkhalek et al.,2022)
		Whole plant	Ethanol (95%)	*Spodoptera frugiperda*	Neonate larva	98 ± 1.38% survival rate	No antifeedent and contact toxicity	-	4 days	(Fu et al.,2020)
12.	*Senna occidentalis* (L) Link. (Fabaceae)	Leaves Stem	Ethanol Hexane (maceration)	*Aedes aegypti*	Larvicidal (IV)	90-100%	- 117.451 ppm 149.698 ppm	-	24 h	(Sharma et al.,2016)
13.	*Xanthium strumarium* L. (Asteraceae)	Leaves and stem	Ethanol Hexane (maceration)	*Aedes aegypti* L.	Larvicidal (IV)		586.185 ppm 460.923 ppm	-	24 h	(Sharma et al.,2016)
14.	*Mimosa pudica* L. (Fabaceae)	Whole body	Fresh juice	*Aedes*	Larvicidal (III)	100% at 8 m*l*	-	-	48hrs	(Rathy et al.,2015)

S.N.	Name of species/family	Part used	Solvent/ Formulation	Insect tested	Biological assay/stage	% mortality	LC_{50}	LC_{90}	Time	References
15.	*Ageratum houstonianum* Mill. (Asteraceae)	Flower Leaves stem	Ethanol	*Culex pipines*	Larvicidal (III)		259.79 ppm 266.85 ppm 306.86 ppm	657.16 ppm 991.71 ppm 1082.13 ppm	24 h	(Hadidy et al.,2022)
16.	*Sphagneticola trilobata* (L.) Pruski (Asteraceae)	Leaves	Hexane, Dichloromethane	*Spodoptera litura* *Spodoptera exigua* *Plutella xylostella L.*	Larvicidal (III) (contact toxicity and anti-feedant activity)	-	- -	- -	24 h	(Junhirun et al.,2018)
			Ethyl acetate				0.88-4.2 µg/larvae (LD_{50})/ 0.27-2.34 mg/ml (FI_{50})	-		
			Methanol (S0xhlet)				-	-		
17.	*Mimosa diplotricha* Sauvalle (Asteraceae)	Leaves Root	Dried powder	*Callosobruchus maculatus* Fabricius	Adult	50% at 2 gm 52%	-	-	96 h	(Uyi et al.,2020)
18.	*Pistia stratoides* L. (Araceae)	Whole plant	95% Ethanol	*Spodoptera frugiperda*	Neonate larva	99 ± 1.38% survival rate	No anti-feedant and contact toxicity.	-	4 days	(Fu et al.,2020)
		Leaves	Ethyl acetate (percolation) Nine fraction	*Annopheles*	Larvicidal	-	14.81 ppm (Fraction E)	-	24 h.	(Ma et al.,2019)

Note: NM: Not mentioned; - : Not studied; EO: essential oil ; FI50= median feeding deterrence; LC50= median lethal concentration; LC90= lethal concentration on which 90% mortality observed; LD50= median lethal dose

Species-wise number of studies

The species-wise number of studies revealed that *Lantana camara* was the most extensively studied IAPs, with 17 case studies. This was followed by *Mesosphaerum sauveleons* (13 studies), *Chromolena odorata* (10 studies), *Ageratum conyzoides* (6 studies), and *Parthenium hysterophorus*, *Pontederia crassipes*, and *Tithonia diversifolia* (each with 4 studies) (Figure2). Other species had two or fewer than two case studies. The strong aroma and noxious nature of *Lantana camara* and *Mesosphaerum sauveolens* likely contribute to their higher number of studies. Out of 30 IAP species in Nepal, only 18 have been studied for their insecticidal potential and eleven species: *Ipomoea carnea* subsp. *fistulosa* (Mart. ex Choisy), *Mikania micrantha* Kunth., *Alternanthera philoxeroides* (Mart.) Griseb., *Myriophyllum aquaticum* (Vell.)Verdc., *Amaranthus spinosus* L., *Senna tora* (L.) Roxb., *Leersia hexandra* Sw., *Oxalis latifolia* Kunth., *Erigeron karvinskianus* DC, *Galinsoga quadriradiata* Ruiz & Pav, *Leucaena leucocephala* (Lam.) de Wit and *Spermacoce alata* Aubl. were not investigated for insecticidal activity. This discrepancy may be due to researcher's preference for focusing on extensively studied medicinal plants, such as *Azadirchta indica*, for insecticidal properties, and prevalence of large number secondary metabolites and bioactive compounds (Kalisz et al., 2021) while common weeds are often neglected.

Parts of the IAPs used in research

Various plant parts such as leaves, stems, flowers, and roots were used singly or in combination to determine their insecticidal potential. Among these, leaves were the most frequently used, accounting

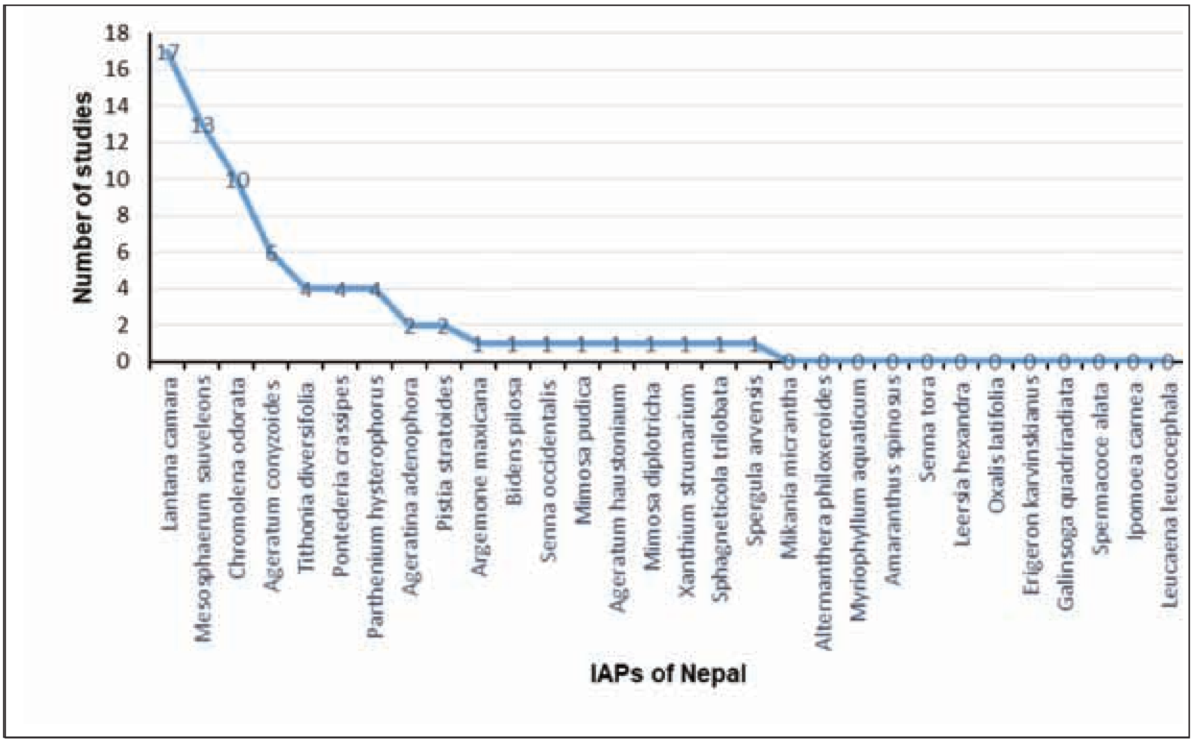

Figure 2: Number of case studies regarding insecticidal potential of different IAPs of Nepal

for more than 50% of the cases, followed by stems, roots, whole plants, and flowers (Figure3). The frequent use of leaves can be attributed to their easy availability in different seasons and their simpler collection process compared to other plant parts. Additionally, the aromatic nature of leaves and presence of more secondary metabolites (Qaderi et al., 2023) may have attracted greater research interest. Among the reviewed cases, leaves were more frequently used part of plant with significant insecticidal effect but there were also evidences for

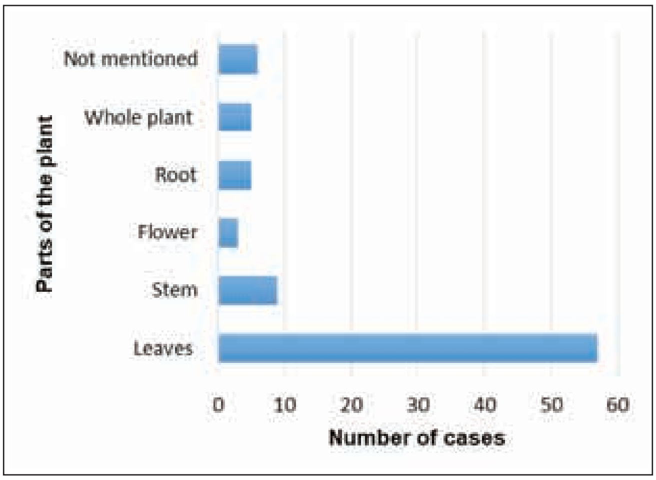

Figure 3: Parts of the IAPs used in published report

the higher insecticidal activity of stem, root and whole-body parts (Milugo et al., 2021). Thus, the review recommends the study of whole-body part of IAPs before their flowering to acquire the benefits for the management of weeds by control and utilization.

Targeted insects and their stages for the study

Among the reported data, 72% cases were studied against the mosquito species belonging to genus *Annopheles*, *Culex* and *Aedes* responsible for malaria, filarial disease, Japanese encephalitis, dengue, etc. whereas 28% were studied against agricultural pests and stored grain pests like *Spodoptera frugiperda*, *Callosobruchus maculatus*, *Spodoptera litura*, *Spodoptera exigua*, *Plutella xylostella*, *Aphis craccivora*, *Atta cephalotes*, *Helicoverpa armigera*, *Pomacea canaliculata*, *Gyraulus convexiusculus*, *Tarebia granifera*, *Tribolium castaneum*, *Lasioderma serricorne*, *Callosobruchus chinensis*, *Sitophilus oryzae*, *Brevicoryne brassicae*, *Hellula undalis*, *Periplanata americana*, *Rhizopertha dominica*, and *Psoroptes lunicula* (Figure 4). Regarding the stages of the targeted insects, larval stages (67%) were used in most of the cases rather than adults (28%). Some cases were also studied for pupae stage (Figure 5).

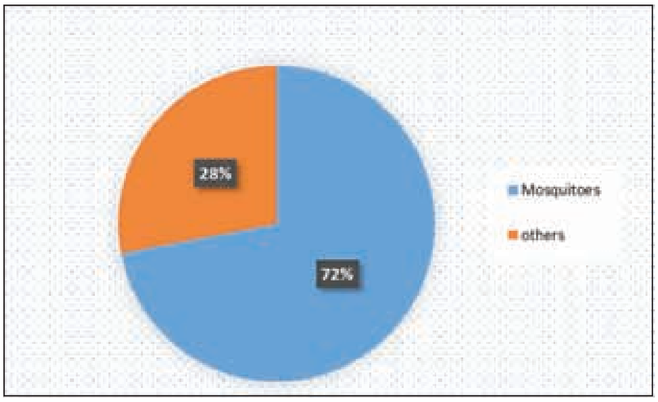

Figure 4*:* Targeted insects for the determination of insecticidal properties of IAPs of Nepal

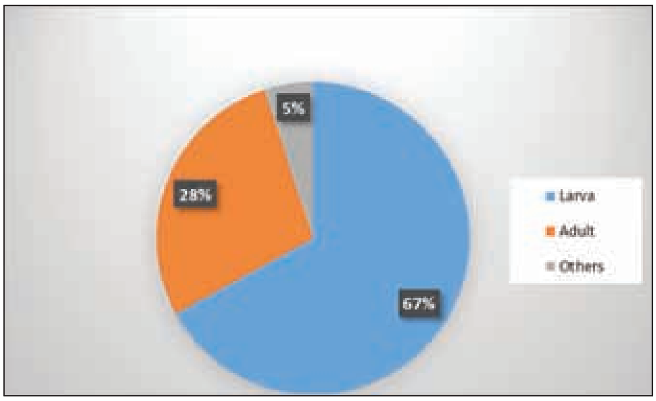

Figure 5: Stages of targeted insects for the determination of insecticidal potential of IAPs

Insecticide formulations

Various methods have been employed to formulate insecticides, including maceration, fresh juice extraction, soxhlet extraction, and hydro-distillation. These methods relied on a range of solvents from polar to non-polar. Methanol (31.66%) was the most commonly used solvent, followed by ethanol (30%). Essential oils obtained through hydro-distillation or steam distillation were also reported to be effective as insecticides. Other solvents and extracts frequently used include hexane and aqueous extracts. Petroleum ether extract was utilized against different insect larvae. Additionally, solvents such as chloroform, acetone, butanol, ethyl acetate, and dichloromethane, as well as fresh juice, and nanoparticles were used in some studies (Table 1).

The efficacy of EO from reviewed cases was found to be stronger with lower LC_{50} and LC_{90} values towards different stages of wide range of insects. This was followed by methanol extract. There were also several evidences for the efficacy of extract formulation of other polar and non-polar solvents as well as the fresh juice. Generally, the evidences showed that petroleum ether extract was more effective to the larval stage than pupa and adult. Other evidences showed efficacy of hexane, chloroform, acetone and aqueous extract but less efficacy of ethanol extract. In order to make cost effective intervention of bio-insecticide, the use of low cost and less harmful solvent is suggested. For this purpose, review recommends the use of higher concentration of aqueous, methanol and petroleum ether extracts as insecticide but for the strongest activity EO can be appropriate. It was found that the effectiveness of the test sample depends on the concentration of the sample and post-exposure periods of sample treatment.

Efficacy of IAPs of Nepal for pest management

Based on the LC_{50} (concentration on which 50% insects were dead) values and/or percentage mortality (Table1), each species was critically analyzed for their insecticidal potential and reported their level of insecticidal effect. Regarding the efficacy of *Lantana camara*, as bio-insecticide for different species of mosquito larva and adult, and agricultural pests, LC_{50} and LD_{50} value was found to be less than 100 ppm post exposure representing higher insecticidal activity. Particularly, EO or EO based nano-emulsion exhibited higher insecticidal potential with lower LC_{50} value than that of the extract formulation with polar and non-polar solvents. Sharma et al. (2021) have reported the larvicidal activity of EO against *Anopheles*, *Aedes* and *Culex* mosquitoes as 94-96% mortality at 100 ppm with LC_{50} (45.32±1.66 ppm) and LC_{90} (96.62±1.99 ppm) at 24hrs. Udappusamy et al. (2022) reported LC_{50} (18.18 ppm- 45.295 ppm) for different larval and pupal stages of *Ades aegypti* and LD50 (11.95 mg/cm^2) for adult when EO loaded nano-emulsion was tested. The EO extracted from different parts of plants collected during rainy and dry season recorded LC_{50} (45 ppm and 41.7 ppm) for larvalstage of *Aedes aegypti* (Luz et al., 2022). According to Aisha et al. (2024), EO on fumigant bioassay/contact toxicity showed LC_{50} (16.70 ppm, 4.141ppm and 6.245ppm) and LD_{50}

(8.93 mg/cm^2, 4.82mg/cm^2, 6.24 mg/cm^2) for the adults of agricultural pests *Tribolium castaneum, Lasioderma serricarne* and *Callosobruchus chinensis*, respectively. The effect of copper nanoparticles isolated from *Lantana* on III instar larva of *Anopheles multicolar* reported to have 100% mortality at 20ppm with LC_{50} 12.5 ppm and LC_{90} 18.4 ppm at 24hrs and compared to petroleum ether extract (LC_{50}= 63.5 ppm and LC_{90}=119.9 ppm), the effect was 5-6 fold higher (Abd El Hafiz Hassanain et al., 2019). Hexane extract was found to be more effective larvicide for *Aedes* with LC_{50} (15.94 ppm and 20.19 ppm) than acetone extract (LC_{50}=32.62 ppm and 106.09 ppm) for sensitive and local strains, respectively. Similarly, petroleum ether extract from leaves of *Lantana* was reported to be more effective to larva of *Aedes* and *Culex* mosquito with the LC_{50} value 74.93 ppm and 10.63 ppm compared to chloroform and methanol extracts (Hari & Mathew, 2018). Thus, *Lantana camara* could be the best alternative of synthetic insecticide for varied insects.

Regarding the efficacy of *Parthenium hysterophorus*, hexane and petroleum ether extract from root was more effective (LC_{50}=87.9 ppm and 10.7 ppm) compared to leaves and stem whereas ethanol extract of all parts showed inferior results against IV in star larva of *Anopheles arabiensis* (Tarekegn et al., 2021). In a study performed by Milugo et al. (2021), the compound parthenin and its two fractions viz ethylene glycol and azide were found to be highly effective against III instar larva of *Anopheles gambiae* with LC_{50} value 154 ppm, 37 ppm and 66 ppm, respectively. They also reported that fraction of the compound parthenin had more insecticidal efficacy than the whole compound. These reported data demonstrate that, not all parts of the plants could have similar insecticidal activity but the level of effect may differ based on their solvent used for extraction, time of collection, methods of application and post exposure duration.

In the case of *Chromolena odorata,* leaves were more frequently studied for insecticidal efficacy than root and stem. In a study carried out in India by Kumar et al. (2023), petroleum ether, chloroform, and ethanol extracts of *Chromolena* against IV instar larva of *Culex* showed strong efficacy with LC_{50} value 0.16 ppm, 0.24 ppm and 0.643 ppm, respectively. However, in the study of Sukhthankar et al. (2014), methanol extract exhibited 100% mortality at 1000 ppm for *Anopheles*, 200ppm for *Culex* and 900 ppm for *Aedes* and the authors argued that methanol extract is highly effective against *Culex* larva than that of other species of mosquitoes. Ileke and Olabimi (2019) reported the efficacy of methanol extract against larva, pupa, and adult of *Anopheles gambiae* with LC_{50} value 31.53 ppm, 53.61ppm and 296.20 ppm, respectively. The methanol extract from leaves of *Chromolena* when tested against tobacco eating *Spodoptera litura,* showed ovicidal (73±0.57%), anti-feedant (82.45±0.16%) and larvicidal (68.33±0.05%) activity at 96 hrs and less mortality was reported in acetone and aqueous extract (Gorawade et al., 2022). Along the wide cases reviewed, effects of test samples also depend upon the species of insects and their stages.

Methanol, hexane, and dichloromethane extracts of whole plant of *Spergula arvensis* were studied against IV in star larva of *Annopheles culicifiacies* for 24 hrs and reported 100% mortality at 40 µg/ml of methanol extract revealing higher efficacy (LC_{50}= 6.84 ppm) than hexane (LC_{50}= 2265.01 ppm) and dichloromethane (LC_{50}= 1951.79 ppm) extracts (Sogan et al., 2021). Similarly, methanol extract from leaves of *Argemone maxicana* showed LC_{50}= 224.45 ppm for IV in star larva of *Anopheles subpictus* in a study by Maheshwari et al. (2023).

Regarding the insecticidal efficacy of *Ageratum conyzoides*, petroleum ether extracted from the leaves of *Ageratum* showed 78%, 77% and 73% mortality of larva of *Aedes*, *Anopheles* and *Culex* mosquitoes with 1.92 ppm, 1.95 ppm and 1.97 ppm (LC_{50}), respectively (Ramasamy et al.,2021) exhibiting the higher level of toxicity compared to other case studies. According to Adelaja et al. (2023), EO from leaves of this plant was effective for *Anopheles* larva (LC_{50}=190 ppm and LC_{90}= 420 ppm) but less effective for the adult. Study of aqueous extract of *Ageratum conyzoides* against pests of cabbage *Plutella xylostella*, *Brevicoryne brassicace, Hellula undalis* and Coccinellidae for

4 weeks reported that habitat manipulation with non-crop plants and spraying contributed to pest suppression along with improved cabbage head yield and quality (Amoabeng et al., 2020). From all of the reported studies, it can be hypothesized that the extract or EO, which is highly effective to one stage of insect, may also be ineffective to the other stage. Additionally, the effect may differ widely among different insects.

Though ethanol extract was reported to be less effective, leaves of *Bidens pilosa* showed 100% larval mortality of *Culex* mosquito at 0.6ml (Ojianwuna et al., 2021). Samuel et al. (2014) studied the potentiality of methanol, chloroform, and petroleum ether extracts from leaves of *Ageratina adenophora* on III instar larva of *Culex* and reported LC_{50} as 509.13 ppm, 335.10 ppm and 186.98 ppm when observed for 24 hrs. Furthermore, Nong et al. (2014) reported 100% adult mortality of *Psoroptes cuniculi* when treated with 50% petroleum ether extract of leaves and stem for 4-6 hrs. Petroleum ether extract exhibited pronounced larvicidal activity compared to other extracts.

Mesosphaerum suaveolens has been reported to be highly effective against different insects attributed to its strong aroma. Obembe et al. (2024) reported 100% larvicidal, pupicidal and adulticidal effects for *Anopheles gambiae* on 0.8, 1 and 0.8% ethanol extracts. EO from *M. suaveolens* had been reported to be highly effective against III and IV instar larva and adult of *Anopheles* with LC_{50} of 3.579 ppm and 6.20 ppm, respectively (Duniya et al., 2022) but LC_{50} was 32.85 ppm for the larva of *Ades aegypti* (Moola et al., 2023) whereas Adelaja et al. (2023) reported LC_{50} (240 ppm) for larva of *Anopheles gambiae*; and larval mortality of *Spodoptera frugiperda* was 6.67±0.5% (de Menezes et al., 2020). Peniche et al. (2022) studied the effect of nano-emulsion from *M. suaveolens*on III instar larva of *Culex quinquefasciatus* and reported 100% mortality at 250 ppm with LC_{50} (102.41 ppm) when observed for 24 hrs. From these cases, it can be concluded that EO or nano-emulsion can also have reduced effects than that of other extracts.

Though, *Tithonia diversifolia* was reported to have 100% mortality of different insects, the LC_{50} value was greater than 1000 ppm which was far higher than other IAPs (Ekemezie,2021). A study by Pantoja-Pulido et al. (2020) on adulticidal effect of leaf cutter ant *Atta cephalotes* with hexane, 70% ethanol, dichloromethane, ethyl acetate butanol and aqueous extracts of *T. diversifolia* reported 70-90% mortality at 1000 ppm dichloromethane extract only within 14 days.

Pontederia crassipes was also reported to have significant insecticidal potential when tested against *Culex* larva and adult of *Aphis craccivora*. The larvae of *Culex pipiens* showed 100% mortality when treated with 75% ethanol and petroleum ether extract from leaves and stem of *Pontederia* whereas petroleum ether extract from root was comparatively more effective than leaves (Hasaballah, 2015). In contrast to all these, 95% ethanol extract from whole plant reported to have 98 ±1.38% survival rate with no anti-feedant and contact toxicity on neonate larva of *Spodoptera frugiperda* (Fu et al.,2020). The effect of *Senna occidentalis*, *Xanthium strumarium*, *Mimosa pudica*, *Ageratum haustonium*, *Sphagneticola trilobata*, *Mimosa diplotricha* and *Pistia stratoides* was also significant to different insects with varied formulations.

This review investigated that the highly effective extract for larval stage can also be least effective to adult stage and vice-versa (Ileke & Olabimi, 2019; Udappusamy et al., 2022; Wangrawa et al., 2023). Also, the evidences presented that the highly effective extract against one type of insect can exhibit moderate to least effects on other types. Furthermore, the lethal dose for targeted insect can also have toxicity to non-target organisms as investigated in the review (Sogan et al., 2021). Hence, the formulation of the insecticide should be insect and stage specific and non-lethal to non-target organisms.

The availability of IAPs in Nepal posed a serious challenge in biodiversity and requires control in their growth and expansion. The potential as an insecticide would promote their use and provide a local/natural solution for insect/pest control.

Different studies were evaluated for this purpose. This review found that most of the IAPs of Nepal could have potentiality in the production of bio-insecticides against wide range of agricultural pests as well as the vector of several human diseases. Based on the above mentioned species-specific evidences representing lower LC_{50} value but higher mortality percentage, it can be concluded that *Lantana camara, Mesosphaerum suaveolens, Chromolena odorata, Ageratina adenophora, Ageratum conyzoides, Parthenium hysterophorus, Bidens pilosa*, *Pontederia crassipes* and *Tithonia diversifolia* could be the potential source of bioactive compound with insecticidal activities. But *Senna occidentalis, Xanthium strumarium, Mimosa pudica, Ageratum haustonium, Sphagneticola trilobata, Mimosa diplotricha* and *Pistia stratoides* need further exploration for insecticidal activity. Though IAPs have negative impact on biodiversity and agriculture resulting in low productivity, this review recommends their utilization for the resource poor farmer to produce cheap, assessable, eco-friendly bio-insecticide instead of using hazardous synthetic insecticides.

Toxicity on non-target organism

Only a few reported studies have investigated the toxicity of plant extracts and EOs on non-target organism. Sogan et al. (2021) found that the methanol extract of *Spergula arvensis* was non-toxic to *Poecilia reticulata* Peters and *Diplonychus indicus* (Fabricius) at concentrations lethal to mosquito larva. Similarly, the petroleum ether extract of *Ageratum conyzoides* was non-toxic to the aquatic predator *Toxorhynchites splendens* (Wiedemann) at 1000 ppm, whereas the commercial pesticide temephos was toxic at 1-2 ppm (Ramasamy et al., 2021). Screening plant extracts from various species, including *Lantana camara* and *M. suaveolens*, against mosquito fish *Gambusia affinins* (Baird & Girard) revealed no mortality at 50 and 60 mg/L. A combination of petroleum ether extract of *Lantana camara*, *Tecoma stans* (L.) Juss.ex Kunth, *M. suaveolens*, and methanol extract of *Nerium oleander* L. in a 1:1:1:1 ratio showed a dose-dependent response above 60 mg/L after 48 hours(Hari &Mathew,2018). Abdelkhalek et al. (2022) reported no deleterious effects or toxicity in albino rats exposed to extracts from *Pontederia crassipes*. However, Luz et al. (2022) advised against using essential oils from all studied plants, including *Lantana camara*, as they were toxic to the non-target organisms *Danio rerio* (Hamilton) and *Artemia salina* (Linnaeus), with high mortality rates even at concentrations below 100µg/mL within few hrs.

Limitations

During the investigation of IAPs of Nepal for their potentiality as insecticides, there were no any published reports from Nepal till date, though concern and studies about management of IAPs is increasing. The IAPs of Nepal which are or aren't invasive to other countries were reported to have been investigated for their insecticidal activity in other countries. The effectiveness of these plants may vary among the geographical range and to the varied insects because of the variation of secondary metabolites on them (Qaderi et al., 2023). The published cases in this review reported to have several limitations and research gaps. Though the preliminary studies had been done to find out the insecticidal potential, most of the published reports lack the investigation regarding bioassay guided fractionation. Some of the studies had presented the chemical data using GC-MS analysis (Pintong et al., 2020; Ramasamy et al., 2021), however, only few studies reported bioactive compound and the insecticidal potential of pure compound and fraction of compounds(Hari & Mathew, 2018; Nong et al., 2014). Similarly, the mode of action of test samples on target organisms was also not investigated in many cases except few studies that have performed acetylcholinesterase (AChE) inhibitor activity of compound (Pantoja-Pulido et al., 2020). The effects of extract and other formulation on non-target organisms was investigated using Albino mice, *Gambusia affinins*, *Danio rerio* (zebrafish), *Artemia salina, Poecilia reticulata* and *Diplonychus indicus* in only five published reports but all other studies lack the test of non-target organisms. All of the studied cases were performed under the laboratory conditions with either wild strain of insects or sensitive strain grown in laboratory but the efficacy under field condition was not reported. While reporting the mortality and lethal concentration, post exposure period was found to be varied ranging between 6 hours to 12 weeks though many cases conducted observation for 24 hours post exposure and the initial and residual activity of extract on target organism was rarely mentioned. The assessment of cross resistance with used bio-insecticides was also not investigated. Similarly, there is gap concerning compatibility with other pest management system; chemical profiling and bioactive compound; and challenges of using IAPs as insecticides. The studies investigated the effect of extract and negative control with solvent used for extract preparation but positive control is lacking. Future studies should assess both positive and negative controls along with the test samples so that the efficacy of the bio-insecticide can be compared with the synthetic insecticide, test conditions can be validated, resistance of insect can be detected and research can be reproducible (Vom Saal & Welshons, 2006). Furthermore, future studies regarding the sustainable management of IAPs should focus towards their utilization for peoples' livelihood and insect eradication with their active participation.

Conclusion

Among the IAPs of Nepal, 18 have been studied for their insecticidal activity and future study can be focus on all 30 IAPs of Nepal. The formulation of test sample and their effectiveness varied across different species, plant parts, and times of collection. Additionally, the insecticidal effect was found to be concentration and time-dependent. EOs and the fraction of extracts were generally more effective than simple extracts. Our study concluded that utilization of IAPs for pest management could be effective against rice weevils, cabbage beetles, stored grain pests, fall armyworms, and varieties of mosquitoes. Future research should focus on identification of bioactive compounds from those IAPs having insecticidal activity. Also the lethal and sub lethal concentration/dose of the extract, fractions and compounds on non-target organism should be done to address the research gap. Research focusing on the comparison of positive and negative control with bio-pesticides can validate the efficacy. Mode of action of the bio-pesticide should be focused. Regarding the environmental safety, the use of non-polar and toxic solvents for the extraction and pesticide formulation should be discouraged but the use of cost-effective, less harmful, polar solvents like methanol, ethanol and water should be encouraged. As the previous researches were more targeted towards larval stage of insects, future prospect could be the study on adulticidal effect of bio-pesticide which will provide immediate solution to problematic insects. In-depth study of IAPs, their chemical profiling, biological activity, bio-active compounds can open up the dual solution of pest management and IAPs management by their utilization.

Author Contributions

T K Chhetri developed the concept and prepared original manuscript, R K Thapa critically analyzed and co-supervised the manuscript and H D Bhattarai reviewed, analyzed and supervised the manuscript preparation.

Acknowledgements

Authors would like to thank Ms. Sushmita Poudel and Ms. Nita Acharya for their insightful suggestions.

References

Abd El Hafiz Hassanain, N., Zeinhom Shehata, A., Mohamed Mokhtar, M., Mohamed Shaapan, R., Abd El Hafiz Hassanain, M., & Zaky, S. (2019). Comparison between insecticidal activity of *Lantana camara* extract and its synthesized nanoparticles against anopheline mosquitoes. *Pakistan Journal of Biological Sciences: PJBS*, *22*(7), 327-334. https://doi.org/10.3923/pjbs.2019.327.334

Abdelkhalek, S. T., Abdelgayed, S. S., Jiang, H., & Wang, M.-Q. (2022). The toxicity of

Eichhornia crassipes fractionated extracts against *Aphis craccivora* and its safety in albino rats. *Toxins*, *14*(5), 327. https://doi.org/10.3390/toxins14050327

Adelaja, O. J., Oduola, A. O., Ande, A. T., Abiodun, O. O., & Adelaja, A. R. (2023). Toxicity of insecticidal plant oils on the larval and adult stages of a major malaria vector (*Anopheles gambiae* Giles 1920). *Parasitology Research*, *122*(5), 1071-1078. https://doi.org/10.1007/s00436-023-07806-6

Afolabi, O. J., Simon-Oke, I. A., Elufisan, O. O., & Oniya, M. O. (2018). Adulticidal and repellent activities of some botanical oils against malaria mosquito: *Anopheles gambiae* (Diptera: Culicidae). *Beni-Suef University Journal of Basic and Applied Sciences*, *7*(1), 135-138. https://doi.org/10.1016/j.bjbas.2017.09.004

Aisha, K., Visakh, N. U., Pathrose, B., Mori, N., Baeshen, R. S., & Shawer, R. (2024). Extraction, chemical composition and insecticidal activities of *Lantana camara* Linn. leaf essential oils against *Tribolium castaneum, Lasioderma serricorne* and *Callosobruchus chinensis*. *Molecules (Basel, Switzerland)*, *29*(2), 344.https://doi.org/10.3390/molecules29020344

Akbar, R., Sun, J., Bo, Y., Khattak, W. A., Khan, A. A., Jin, C., Zeb, U., Ullah, N., Abbas, A., Liu, W., Wang, X., Khan, S. M., & Du, D. (2024). Understanding the influence of secondary metabolites in plant invasion strategies: A comprehensive review. *Plants*, *13*(22), 3162. https://doi.org/10.3390/plants13223162

Amoabeng, B. W., Stevenson, P. C., Mochiah, B. M., Asare, K. P., & Gurr, G. M. (2020). Scope for non-crop plants to promote conservation biological control of crop pests and serve as sources of botanical insecticides. *Scientific Reports*, *10*(1), 6951. https://doi.org/10.1038/s41598-020-63709-x

Annie, S. W., Raveen, R., Paulraj, M. G., Samuel, T., & Arivoli, S. (2015). Screening of *Eichhornia crassipes* (Mart.) Solms (Pontederiaceae) crude leaf extracts for larvicidal efficacy against the filarial vector *Culex quinquefasciatus* say (Diptera: Culicidae). *International Journal of Mosquito Research*, *2*, 43-48. https://www.dipterajournal.com/vol2issue4/pdf/2-3-13.1.pdf

Aremu, H. K., Adekale, I. A., Azeez, L. A., Busari, H. K., Adebisi, O., Iwalewa, Z. O., Alle, O. E., & Musa, D. A. (2022). Assessment of larvicidal and genotoxic potentials of extracts of *Hyptis suaveolens* against *Culex quinquefasciatus* based on enzyme profile and RAPD-PCR assay. *Acta Tropica*, *229*, 106384. https://doi.org/10.1016/j.actatropica.2022.106384

Ascher, K. R. S. (1993). Nonconventional insecticidal effects of pesticides available from the Neem tree, *Azadirachta indica*. *Archives of Insect Biochemistry and Physiology*, *22*(3-4), 433-449. https://doi.org/10.1002/arch.940220311

Ayilara, M. S., Adeleke, B. S., Akinola, S. A., Fayose, C. A., Adeyemi, U. T., Gbadegesin, L. A., Omole, R. K., Johnson, R. M., Uthman, Q. O., & Babalola, O. O. (2023). Biopesticides as a promising alternative to synthetic pesticides: A case for microbial pesticides, phytopesticides, and nanobiopesticides. *Frontiers in Microbiology*, *14*, 1040901. https://doi.org/10.3389/fmicb.2023.1040901

Bini, K. K. N., Kobenan, K. C., Kouakou, M., Kouadio, I. S., Zengin, G., Jekő, J., Cziáky, Z., Danho, M., & Ochou, O. G. (2023). Phytochemical profiling, antioxidant activities, enzymatic activities and insecticidal potential of aqueous extracts of four plants on the larvae of *Helicoverpa armigera* (Lepidoptera: Noctuidae), the main pest of cotton plant in Ivory Coast. *Archives of Insect Biochemistry and Physiology*, *113*(3), e22017. https://doi.org/10.1002/arch.22017

Chaudhary, S., Kanwar, R. K., Sehgal, A., Cahill, D. M., Barrow, C. J., Sehgal, R., & Kanwar, J. R. (2017). Progress on *Azadirachta indica* based biopesticides in replacing synthetic toxic pesticides. *Frontiers in Plant Science*, *8*. https://doi.org/10.3389/fpls.2017.00610 de Menezes, C. W. G., Carvalho, G. A., Alves, D. S., de Carvalho, A. A., Aazza, S., de Oliveira Ramos, V., Pinto, J. E. B. P., & Bertolucci, S. K. V. (2020). Biocontrol potential of methyl chavicol for managing *Spodoptera frugiperda* (Lepidoptera: Noctuidae), an important corn pest. *Environmental Science and Pollution Research International*, *27*(5), 5030-5041.https://doi.org/10.1007/s11356-019-07079-6

Duniya, S. V., Joseph, B. C., & Sokoato, M. I. (2022). Insecticidal and mosquito repellency property of essential oil from *hyptis suaveolens* leaves. *Open Access Journal of Science*, *5*(1), 77-83. https://doi.org/10.15406/oajs.2022.05.00179

Ekemezie, S. (2021). Activity of *Lantana camara* Linn. (Wide Sage) and *Tithonia diversifolia* (Hemsl) A. Gray (Mexican Sunflower) leaf extract on *Anopheles* mosquito larvae. *American Journal of Infectious Diseases and Microbiology*, *9*, 10-17. https://doi.org/10.12691/ajidm-9-1-3

Elemike, E. E., Onwudiwe, D. C., Ekennia, A. C., Sonde, C. U., & Ehiri, R. C. (2017). Green synthesis of Ag/Ag, O nanoparticles using aqueous leaf extract of *Eupatorium odoratum* and its antimicrobial and mosquito larvicidal activities. *Molecules (Basel, Switzerland)*, *22*(5), 674. https://doi.org/10.3390/molecules22050674

Essiedu, J. A., Adepoju, F. O., & Ivantsova, M. N. (2020). Benefits and limitations in using biopesticides: A review. *AIP Conference Proceedings*, *2313*(1), 080002. https://doi.org/10.1063/5.0032223

Eze, A., Gloria, O., Ikuesan, A., & Belllo, A. (2022). Larvicidal synergistic efficacy of plant parts of *Lantana camara* against *Aedes aegypti*. *Journal of entomology and zoology studies*, *10*, 187-192. https://doi.org/10.22271/j.ento.2022.v10.i1c.8934

Fu, Y., Bhadha, J. H., Rott, P., Beuzelin, J. M., & Kanissery, R. (2020). Investigating the use of aquatic weeds as biopesticides towards promoting sustainable agriculture. *PLoS ONE*, *15*(8), e0237258. https://doi.org/10.1371/journal.pone.0237258

Gorawade, V., Attar, U., & Shiragave, P. (2022). *Bioefficacy and GC-MS analysis of Chromolaena odorata and Leonotis nepetifolia* leaf extracts against *spodoptera litura*. *Journal of Crop Protection*, *11*(3), 361-375. https://doaj-org.tucl.remotexs.co/article/e5c412099b904abdb9a2892a9eb77a62

Hadidy, D. E., El Sayed, A. M., Tantawy, M. E., Alfy, T. E., Farag, S. M., & Haleem, D. R. A. (2022). Larvicidal and repellent potential of *Ageratum houstonianum* against *Culex pipiens*. *Scientific Reports*, *12*(1), 21410. https://doi.org/10.1038/s41598-022-25939-z

Hari, I., & Mathew, N. (2018). Larvicidal activity of selected plant extracts and their combination against the mosquito vectors *Culex quinquefasciatus* and *Aedes aegypti*. *Environmental Science and Pollution Research*, *25*(9), 9176-9185.https://doi.org/10.1007/s11356-018-1515-3

Hasaballah, A. I. (2015). Toxicity of some plant extracts against vector of lymphatic filariasis, *Culex pipiens*. *Journal of the Egyptian Society of Parasitology*, *45*(1), 183-192. https://doi.org/10.12816/0010864

Hicks, H. L., Comont, D., Coutts, S. R., Crook, L., Hull, R., Norris, K., Neve, P., Childs, D. Z., & Freckleton, R. P. (2018). The factors driving evolved herbicide resistance at a national scale. *Nature Ecology & Evolution*, *2*(3), 529-536. https://doi.org/10.1038/s41559-018-0470-1

Hiremath, G. I., Ahn, Y.-J., & Kim, S.-I. (1997). Insecticidal activity of Indian plant extracts against *Nilaparvata lugens* (Homoptera: Delphacidae). *Applied Entomology and Zoology*, *32*(1), 159-166. https://doi.org/10.1303/aez.32.159

Huy Hung, N., Ngoc Dai, D., Satyal, P., Thi Huong, L., Thi Chinh, B., Quang Hung, D., Anh Tai, T., & Setzer, W. N. (2021). *Lantana camara* essential oils from Vietnam: Chemical composition, molluscicidal, and mosquito larvicidal activity. *Chemistry & Biodiversity*, *18*(5), e2100145. https://doi.org/10.1002/cbdv.202100145

Ileke Kayode, D., & Olabimi Isaac, O. (2019). Insecticidal activities of *Chromolaena odorata* and *Vernonia amygdalina* leaf extracts against *Anopheles gambiae* [Diptera: Culicidae]. *International Journal of Tropical Diseases*, *2*(1). https://doi.org/10.23937/ijtd-2017/1710018

Imam, A. A., Amos, A. S., Abdullahi, H., & Ibrahim, A. (2018). Comparative larvicidal properties and detoxification machinery of bioactive fractions of leaf extracts of *Hyptis Suaveolens* and *Chromolaena Odorata* on *Anopheles gambiae* s.l from North West Nigeria. *Malaysian Journal of Applied Sciences*, *3*(1), 9-23. https://journal.unisza.edu.my/myjas/index.php/myjas/article/view/80

Jayaraman, T., Chandrapragasam, V., & Subbiah, K. (2023). Development of bioformulations using plant extracts for the control of dengue vector, *Aedes aegypti*. *Journal of Applied and Natural Science*, *15*(2), Article 2. https://doi.org/10.31018/jans.v15i2.4518

Junhirun, P., Pluempanupat, W., Yooboon, T., Ruttanaphan, T., Koul, O., & Bullangpoti, V. (2018). The study of isolated alkane compounds and crude extracts from *Sphagneticola trilobata* (Asterales: Asteraceae) as a candidate botanical insecticide for Lepidopteran larvae. *Journal of Economic Entomology*, *111*(6), 2699-2705. https://doi.org/10.1093/jee/toy246

Kalisz, S., Kivlin, S. N., & Bialic-Murphy, L. (2021). Allelopathy is pervasive in invasive plants. *Biological Invasions*, *23*(2), 367-371. https://doi.org/10.1007/s10530-020-02383-6

Kandalkar, A., Kshirsagar, S., Jundare, P., & Jaiswar, S. (2020). Study of *Lantana Camara*, *Parthenium Hysterophorus* and *Allium Sativum* Linn. for mosquito larvicidal potential and the formulation of larvicidal Sachet. *American Journal of PharmTtech Research*, *10*(4), 019. https://doi.org/10.46624/ajptr.2020.v10.i4.019

Kumar, J., Ramlal, A., Mallick, D., & Mishra, V. (2021). An overview of some biopesticides and their importance in plant protection for commercial acceptance. *Plants*, *10*(6), 1185. https://doi.org/10.3390/plants10061185

Kumar, R. A., Kowsalya, C., & Jeyasankar, A. (2023). Preliminary phytochemical analysis and larvicidal property of *Chromolaena odorata* (L.) against *Culex quinquefasciatus*. *International Journal of Mosquito Research*, *10*(5), 35-39. https://doi.org/10.22271/23487941.2023.v10.i5a.694

Luz, T. R. S. A., Leite, J. A. C., de Mesquita, L. S. S., Bezerra, S. A., Gomes Ribeiro, E. C., Silveira, D. P. B., Mesquita, J. W. C. de, do Amaral, F. M. M., & Coutinho, D. F. (2022). Seasonal variation in the chemical composition and larvicidal activity against *Aedes aegypti* L. of essential oils from Brazilian Amazon. *Experimental Parasitology*, *243*, 108405. https://doi.org/10.1016/j.exppara.2022.108405

Ma, J., Ugya, Y. A., Isiyaku, A., Hua, X., & Imam, T. S. (2019). Evaluation of *Pistia stratiotes* fractions as effective larvicide against *Anopheles* mosquitoes. *Artificial Cells, Nanomedicine, and Biotechnology*, *47*(1), 945-950. https://doi.org/10.1080/21691401.2019.1582538

Macel, M., de Vos, R. C. H., Jansen, J. J., van der Putten, W. H., & van Dam, N. M. (2014). Novel chemistry of invasive plants: Exotic species have more unique metabolomic profiles than native congeners. *Ecology and Evolution*, *4*(13), 2777-2786. https://doi.org/10.1002/ece3.1132

Maheshwari, R., Maddala, M., Lingakari, M., & Mayookha, L. (2023). Natural mosquito control: *Vitex negundo and Argemone mexicana* as larvicidal agents against *Anopheles subpictus*. *International Journal of Mosquito Research*, *10*, 15-19. https://doi.org/10.22271/23487941.2023.v10.i4a.681

Milugo, T. K., Tchouassi, D. P., Kavishe, R. A., Dinglasan, R. R., & Torto, B. (2021). Derivatization increases mosquito larvicidal activity of the sesquiterpene lactone parthenin isolated from the invasive weed *Parthenium hysterophorus*. *Pest Management Science*, *77*(2), 659-665. https://doi.org/10.1002/ps.6131

Moola, A. K., Ayyadurai, T., Balasubramani, S., Vignesh, R., Mohan, P. K., Sathish, S., & Diana, R. K. B. (2023). Chemical composition and larvicidal activity against *Aedes aegypti* larvae from *Hyptis suaveolens* (L.) Poit essential oil. *Journal of Natural Pesticide Research, 3*, 100018. https://doi.org/10.1016/j.napere.2022.100018

Nenaah, G. E. (2014). Bioactivity of powders and essential oils of three Asteraceae plants as post-harvest grain protectants against three major coleopteran pests. *Journal of Asia-Pacific Entomology*, *17*(4), 701-709. https://doi.org/10.1016/j.aspen.2014.07.003

Nong, X., Li, S.-H., Chen, F.-Z., Wang, J.-H., Xie, Y., Fang, C.-L., Liu, T.-F., He, R., Gu, X.-B., Peng, X.-R., & Yang, G.-Y. (2014). Isolation and identification of acaricidal compounds in *Eupatorium adenophorum* petroleum ether extract and determination of their acaricidal activity against *Psoroptes cuniculi*. *Veterinary Parasitology*, *203*(1-2), 197-202. https://doi.org/10.1016/j.vetpar.2014.01.029

Obembe, O. M., Omotoso, O. T., & Olorunniyi, O. F. (2024). Insecticidal potential of *Eucalyptus citriodora* Hook and *Hyptis suavolens* (L.) Poit ethanol leaves extracts against malaria mosquito (*Anopheles gambiae* Giles). *Asian Journal of Advanced Research and Reports*, *18*(5), 72-79. https://doi.org/10.9734/ajarr/2024/v18i5633

Ojianwuna, C. C., Ilondu, E. M., & Enwemime, V. N. (2021). Larvicidal efficacy of leaf extracts from three asteraceous plant against mosquito (*Culex quinquefasciatus*). *Fudma journal of sciences*, *5*(2), 100-108. https://doi.org/10.33003/fjs-2021-0502-528

Oluwamodupe, C., Sb, S., Olayeriju, O. S., & Suresh, K. (2024). Phytochemical characterisation and toxicity effect of *Tithonia diversifolia* (Hemls.) A. Gray leaf extract on fall army worm *Spodoptera frugiperda* (JE Smith) larvae. *Journal of Complementary & Integrative Medicine*. *21*(3). https://doi.org/10.1515/jcim-2023-0310

Pandey, M., Magar, K. B. T., Poudel, B. S., Davis, T. S., & Shrestha, B. B. (2020). Plant invasion research in Nepal: A review of recent national trends. *Weeds-Journal of the Asian-Pacific Weed Science Society*, *2*(2), 16-33.

Pantoja-Pulido, K. D., Rodríguez, J., Isaza-Martínez, J. H., Gutiérrez-Cabrera, M., Colmenares-Dulcey, A. J., & Montoya-Lerma, J. (2020). Insecticidal and cholinesterase activity of dichloromethane extracts of *Tithonia diversifolia* on *Atta cephalotes* worker ants (formicidae: myrmicinae). *Insects*, *11*(3), 180. https://doi.org/10.3390/insects11030180

Peniche, T., Duarte, J. L., Ferreira, R. M. A., Sidônio, I. A. P., Sarquis, R. S. F. R., Sarquis, Í. R., Oliveira, A. E. M. F. M., Cruz, R. A. S., Ferreira, I. M., Florentino, A. C., Carvalho, J. C. T., Souto, R. N. P., & Fernandes, C. P. (2022). Larvicidal effect of *Hyptis suaveolens* (L.)Poit. essential oil nanoemulsion on *Culex quinquefasciatus* (Diptera: Culicidae). *Molecules (Basel, Switzerland)*, *27*(23), 8433. https://doi.org/10.3390/molecules27238433

Pimentel, D., McNair, S., Janecka, J., Wightman, J., Simmonds, C., O'connell, C., Wong, E., Russel, L., Zern, J., & Aquino, T. (2001). Economic and environmental threats of alien plant, animal, and microbe invasions. *Agriculture, Ecosystems & Environment*, *84*(1), 1-1-20. https://doi.org/10.1016/S0167-8809(00)00178-X

Pintong, A., Ampawong, S., Komalamisra, N., Sriwichai, P., Popruk, S., & Ruangsittichai, J. (2020). Insecticidal and histopathological effects of *Ageratum conyzoides* weed extracts against dengue vector, *Aedes aegypti*. *Insects*, *11*(4), 224. https://doi.org/10.3390/insects11040224

Qaderi, M. M., Martel, A. B., & Strugnell, C. A. (2023). Environmental factors regulate plant secondary metabolites. *Plants*, *12*(3), 447. https://doi.org/10.3390/plants12030447

Rajashekar, Y., Ravindra, K. V., & Bakthavatsalam, N. (2014). Leaves of *Lantana camara* Linn.

(Verbenaceae) as a potential insecticide for the management of three species of stored grain insect pests. *Journal of Food Science and Technology*, *51*(11), 3494-3499. https://doi.org/10.1007/s13197-012-0884-8

Ramasamy, V., Karthi, S., Ganesan, R., Prakash, P., Senthil-Nathan, S., Umavathi, S., Krutmuang, P., & Vasantha-Srinivasan, P. (2021). Chemical characterization of billy goat weed extracts *Ageratum conyzoides* (Asteraceae) and their mosquitocidal activity against three blood-sucking pests and their non-toxicity against aquatic predators. *Environmental Science and Pollution Research*, *28*(22), 28456-28469. https://doi.org/10.1007/s11356-021-12362-6

Rathy, M. C., Sajith, U., & Harilal, C. C. (2015). Plant diversity for mosquito control: A preliminary study. *International Journal of Mosquito Research*, *2*(1), 29-33. https://www.dipterajournal.com/vol2issue1/2-1-6.1.html

Rodríguez, J., Montoya-Lerma, J., & Calle, Z. (2015). Effect of *Tithonia diversifolia* mulch on *Atta cephalotes* (Hymenoptera: Formicidae) nests. *Journal of Insect Science (Online)*, *15*(1), 32. https://doi.org/10.1093/jisesa/iev015

Roy, H. E., Pauchard, A., Stoett, P. J., Renard Truong, T., Meyerson, L. A., Bacher, S., Galil, B. S., Hulme, P. E., Ikeda, T., Kavileveettil, S., McGeoch, M. A., Nuñez, M. A., Ordonez, A., Rahlao, S. J., Schwindt, E., Seebens, H., Sheppard, A. W., Vandvik, V., Aleksanyan, A., ... & Ziller, S. R. (2024). Curbing the major and growing threats from invasive alien species is urgent and achievable. *Nature Ecology & Evolution*, *8*(7), 1216-1223. https://doi.org/10.1038/s41559-024-02412-w

Samuel, L., Lalrotluanga, Muthukumaran, R. B., Gurusubramanian, G., & Senthilkumar, N. (2014). Larvicidal activity of *Ipomoea cairica* (L.) Sweet and *Ageratina adenophora* (Spreng.) King & H. Rob. plant extracts against arboviral and filarial vector, *Culex quinquefasciatus* Say (Diptera: Culicidae). *Experimental Parasitology*, *141*, 112-121. https://doi.org/10.1016/j.exppara.2014.03.020

Sani, F., Mairiga, J. P., & Hassan, S. C. (2021). Bio-Potency of *Hyptis suaveolens* Leaf on the Larvicidal Stages of Development of *Anopheles Mosquitoes*. *Lapai Journal of Applied and Natural Science*, *5*(1), 213-219. https://www.lajans.com.ng/article.php?id=135

Sasan, J. M., Lumantao, R. M., Magallon, C. L., Canillo, N. M., Rosalita, E., & Magallon, M. A. (2021). Botanical potency of *Chromolaena odorata* linn (Hagonoy) as mosquitocidal. *Science and Education*, *2*(11), 168-177. https://doi.org/10.52403/ijshr.20211033

Şengül Demirak, M. Ş., & Canpolat, E. (2022). Plant-based bioinsecticides for mosquito control: Impact on insecticide resistance and disease transmission. *Insects*, *13*(2), 8-24. https://doi.org/10.3390/insects13020162

Sharma, A., Kumar, S., & Tripathi, P. (2016). Evaluation of the larvicidal efficacy of five indigenous against an Indian strain of Dengue Vector, *Aedes aegypti* L. (Diptera: Culicidae). *Journal of Parasitology Research*, *2016*, e2857089. https://doi.org/10.1155/2016/2857089

Sharma, M., Alexander, A., Nakhate, K. T., Nagwanshi, K. K., & Ajazuddin, null. (2023). Evaluation of the mosquito larvicidal potential and comparative assessment of the juice of *Lantana camara* Linn and *Ocimum gratissimum* Linn. *Experimental Parasitology*, *249*, 108521. https://doi.org/10.1016/j.exppara.2023.108521

Sharma, M., Alexander, A., Saraf, S., Saraf, S., Vishwakarma, U. K., Nakhate, K. T., & Ajazuddin. (2021). Mosquito repellent and larvicidal perspectives of weeds *Lantana camara* L. and *Ocimum gratissimum* L. found in Central India. *Biocatalysis and Agricultural Biotechnology*, *34*, 102040. https://doi.org/10.1016/j.bcab.2021.102040

Shrestha, B. B. (2016). Invasive alien plant species in Nepal. *Frontiers of Botany*, *2016*, 269-284.

Shrestha, B. B., & Shrestha, K. K. (2021). Invasions of alien plant species in Nepal: patterns and process. In T. Pullaiah, & M. R. Ielmini (Eds.), *Invasive Alien Species* (1st ed., pp. 168-183). Wiley. https://doi.org/10.1002/9781119607045.ch20

Shrestha, B. B., Poudel, A. S., & Pandey, M. (2024). Plant Invasions in Nepal: What we do not know? In M. B. Rokaya, & S. R. Sigdel (Eds.), *Flora and Vegetation of Nepal* (pp. 333-360). Springer International Publishing. https://doi.org/10.1007/978-3-031-50702-1_13

Shrestha, U. B., & Shrestha, B. B. (2019). Climate change amplifies plant invasion hotspots in Nepal. *Diversity and Distributions, 25*(10), 1599-1612. https://doi.org/10.1111/ddi.12963

Sogan, N., Kala, S., Kapoor, N., & Nagpal, B. N. (2021).Phytochemical analysis of *Spergula arvensis* and evaluation of its larvicidal activity against malarial vector *An.culicfiacies*. *South African Journal of Botany*, *137*, 351-358. https://doi.org/10.1016/j.sajb.2020.11.006

Sparks, T. C., & Nauen, R. (2015). IRAC: Mode of action classification and insecticide resistance management. *Pesticide Biochemistry and Physiology*, *121*, 122-128. https://doi.org/10.1016/j.pestbp.2014.11.014

Sukhthankar, J. H., Kumar, H., Godinho, M. H. S., & Kumar, A. (2014). Larvicidal activity of methanolic leaf extracts of plant, *Chromolaena odorata* L. (Asteraceae) against vector mosquitoes. *International Journal of Mosquito Research*, *1*(3), 33-38. https://www.dipterajournal.com/pdf/2014/vol1issue3/PartA/16.1-735.pdf

Tarekegn, M., Wolde-hawariat, Y., Dugassa, S., & Tekie, H. (2021). Evaluation of larvicidal activities of *Parthenium hysterophorus* L. against *Anopheles arabiensis* (Diptera: Culicidae), the major malaria vector in Ethiopia. *International Journal of Tropical Insect Science*, *41*(2), 1461-1469. https://doi.org/10.1007/s42690-020-00344-z

Tawfeek, M. E., Ali, H. M., Akrami, M., & Salem, M. Z. M. (2021).Potential insecticidal activity of four essential oils against the rice weevil, Sitophilus oryzae (L.) (Coleoptera: Curculionidae). *BioResources*, *16*(4), 7767-7783. https://doi.org/10.15376/biores.16.4.7767-7783

Tlak Gajger, I., & Dar, S. A. (2021). Plant Allelochemicals as sources of insecticides. *Insects*, *12*(3), 189. https://doi.org/10.3390/insects12030189

Udappusamy, V., Mohan, H., & Thinagaran, R. (2022). *Lantana camara* L. essential oil mediated nano-emulsion formulation for biocontrol application: Anti-mosquitocidal, anti-microbial and antioxidant assay. *Archives of Microbiology*, *204*(7), 388. https://doi.org/10.1007/s00203-022-02975-x

Udebuani, A. C., Abara, P. C., Obasi, K. O., & Okuh, S. U. (2015). Studies on the insecticidal properties of *Chromolaena odorata* (Asteraceae) against adult stage of *Periplaneta americana*. *Journal of Entomology and Zoology Studies*, *3*(1), 318-321. https://www.entomoljournal.com/archives/?year=2015&vol=3&issue=1&ArticleId=138

Uyi, O. O., Udeogwu, C. C., & Rotimi, J. (2020). Phytochemical constituents and insecticidal efficacy of the root and leaf powders of *Mimosa diplotricha* and *Aspilia africana* against *Callosobruchus maculates* (Fab.) (Coleoptera: Chrysomelidae). *Journal of Applied Sciences and Environmental Management*, *24*(4). https://doi.org/10.4314/jasem.v24i4.16

Vom Saal, F. S., & Welshons, W. V. (2006).Large effects from small exposures. II. The importance of positive controls in low-dose research on bisphenol A. *Environmental Research*, *100*(1), 50-76.

Wangrawa, D. W., Badolo, A., Guenne, S., Guelbéogo, W. M., Kiendrébeogo, M., Sagnon, N., & Sanon, A. (2016). Larvicidal and oviposition-deterrence activities of four local plant extracts from Burkina Faso against *Anopheles gambiae S. l.* (Diptera: Culicidae). *International Journal* *of Mosquito Research*, *3*(6), 11-19. https://www.dipterajournal.com/pdf/2016/vol3issue6/PartA/3-4-17-558.pdf

Wangrawa, D. W., Yaméogo, F., Sombié, A., Esalimba, E., Ochomo, E., Borovsky, D., Badolo, A., & Sanon, A. (2023). Methanol and acetone extracts from the leaves of selected aromatic plants affect survival of field collected *Anopheles arabiensis* (Diptera: Culicidae) from Kisumu, Kenya. *Journal of Medical Entomology*, *60*(5), 1030-1037. https://doi.org/10.1093/jme/tjad066

Williams, L. A. D., & Mansingh, A. (1993). Pesticidal potentials of tropical plants-I. Insecticidal activity in leaf extracts of sixty plants. *International Journal of Tropical Insect Science*, *14*(5-6), 697-700. https://doi.org/10.1017/S1742758400018142

Yadav, R., Tyagi, V., Tikar, S. N., Sharma, A. K., Mendki, M. J., Jain, A. K., & Sukumaran, D. (2014). Differential larval toxicity and oviposition altering activity of some indigenous plant extracts against dengue and chikungunya vector *Aedes albopictus*. *Journal of Arthropod-Borne Diseases*, *8*(2), 174-185. https://jad.tums.ac.ir/index.php/jad/article/view/216

Journal of Plant Resources (2025) **23**(1), 131-155
https://doi.org/10.3126/bdpr.v23i1.83359

Review Article

Current Outlook of Ethnobotany in Bagmati Province, Central Nepal

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Abstract

This paper reviews the state and prospects of ethnobotany in Bagmati Province, central Nepal, based on 102 studies identified through systematic searches using keywords like 'ethnobotany', 'traditional knowledge', and 'indigenous knowledge'. The research employs a forward-backward reference searching approach to analyze 2,245 use reports for 557 plant species from 139 families, categorized into 15 use categories. The Fabaceae family was the most diverse (43 species), followed by Asteraceae (33 species) and Lamiaceae (27 species). Traditional medicine dominates plant use, accounting for 1,822 use reports. Regions such as Makwanpur (270 species), Sindhuli (149 species), and Dolakha (127 species) showed higher species diversity due to extensive field studies and ecological variation from subtropical lowlands to mid-hills. In contrast, high-altitude (Rasuwa, Sindhupalchok) and urbanized districts (Kathmandu) recorded fewer medicinal plants due to sparse populations and alternative healthcare access. Ethnicity-wise, the Newar (327) and Tamang (257) groups recorded the highest level of species diversity. The Brahmin and Chhetri groups, following a similar pattern, recorded 242 species However, Gurung, Pahari, Sarki, and Chepang recorded the least species. The larger ethnic groups documenting extensive usage and smaller groups preserving unique knowledge play an indispensable role in biodiversity conservation. This underscores the inter-relationship between ecological diversity, ethnic group distribution, and ethnobotanical knowledge. Integrating this traditional knowledge into national policies is important for conserving indigenous plant resources and fostering sustainability. Comprehensive surveys in underexplored areas, strengthening documentation, accreditation of traditional healers, and policy frameworks for drug development and quality assurance are vital for promoting ethnobotanical knowledge and its applications.

Keywords: Economic botany, Ethnic group, Indigenous knowledge, Mountains, Traditional medicine

Introduction

Ethnobotany, connecting natural and social sciences, investigates the relationship between people and plants. Of all plant applications, those related to human health and well-being stand out as the most diverse. Plant-based traditional medicine has been widely used around the world, with growing evidence highlighting its importance for both treating diseases and supporting overall health. The secondary metabolites found in medicinal plants provide effective physiological responses, help in relieving human suffering, and are extensively used for subsistence and in treating different illnesses at home (Giday et al., 2009; Kunwar, 2006). The systematic study of these plants offers valuable insights into the ways indigenous communities are involved with natural resources, particularly in medicine and pharmaceutical development (Albuquerque & Hanazaki, 2009). Plant resources are still utilized to meet basic needs or to sell as edible fruits, vegetables, fodder, medicinal herbs, and raw materials forbuilding and household goods in many parts of Nepal (Dulal et al., 2022).

In recent decades, ethnobotany has gained greater recognition as it offers insights into biodiversity conservation efforts and preserves the cultural heritage of Indigenous communities. Several researchers have documented plant-based traditional knowledge across various cultures and regions, making significant contributions to the field of ethnobotany. In addition, ethnomedicinal studies have been instrumental in discovering natural and synthetic drugs such as reserpine, aspirin, taxol etc and in recent years, ethnobotanical insights have continued to support successful drug screening programs (Fabricant & Farnsworth, 2001; Heinrich

& Bremner, 2006). According to the World Health Organization (WHO), approximately 80% of the global population, particularly rural communities in developing countries, still depend on traditional medicines (Islam, 2006). Additionally, more than 50% of all pharmaceutical drugs have origins rooted in ethnomedicine (Wyk et al., 1997).

The variations in physiographic characteristics and climate have contributed to Nepal's enormous diversity of flora, fauna, ecosystems, and cultural heritage (Chaudhary et al., 2020). The Nepal Himalayas are home to a wide variety of plant resources and current flora records comprise roughly 6,000 flowering plant species (https://floraofnepal.org.np/). Of these, between 1,792 and 2,331 species are medicinal and aromatic plants (MAPs), used traditionally by local communities as medicines (Baral & Kurmi, 2006; Rokaya et al., 2010). On the other hand, the country's rich cultural and religious diversity supports a longstanding tradition of herbal medicine. There exists a deeply rooted and extensive history of traditional knowledge regarding medicinal and therapeutic herbs in Nepal (Kunwar & Bussmann, 2008). The indigenous peoples, with a unique culture, identity, and traditions, have occupied the country's diverse geographical and ecological regions. Across the country, they maintain a deep social, cultural, economic, and spiritual bond with natural resources and have developed their own traditional healing and medicinal practices. Acknowledging its diverse heritage, Nepal is home to 60 legally recognized Indigenous Nationalities, who comprise approximately 29.2 percent of the total national population (National Statistics Office [NSO], 2023). These people are profoundly associated with natural resources and maintain an insightful social, cultural, economic, and spiritual connection to these resources.

While numerous ethnobotanical studies have recorded various plants used by different ethnic groups in Central Nepal (Bhattarai, 1989, 1990; Chhetri & Gauchan, 2007; Joshi et al., 2011; Kunwar et al., 2006; Kunwar & Bussmann, 2008; Luitel et al., 2014; Malla & Chhetri, 2009; Manandhar, 1991; Pratik et al., 2011; Rijal, 2008; Shah & Lamichhane, 2017; Shrestha & Dhillion, 2003; Tamang, 2003; Thapa, 2012). Still a considerable amount of information about various plant species remains unexplored and in need of documentation. Furthermore, these fragmented pieces of information need to be reviewed, synthesized, and combined into a single document.

This study presents an in-depth systematic review of published literature on plants utilized by various ethnic communities residing in Bagmati Province, Central Nepal. This review aims to provide a comprehensive overview of the ethnobotanical use of plants.

Materials and Methods

Study area

Bagmati province lies in Central Nepal consisting of 13 districts such as Rasuwa, Sindhupalchok, Dolakha, Dhading, Nuwakot, Kathmandu, Bhaktapur, Lalitpur, Kabhrepalanchok, Ramechhap, Chitwan, Makawanpur and Sindhuli (Figure 1). It has an area of 20,300 sq.Km (13.79% of the total area of the country), of which 27.29% of the land is covered by forest (Bagmati Province Government [BPG], 2024).

Review of literature

An extensive review of the literature was conducted on ethnobotany, traditional knowledge, and indigenous practices in Bagmati Province, Central Nepal. The uses of plants of particular ethnic groups in a particular district or a village were reviewed (Lesnikowski et al., 2013). The general ethnobotanical publications without district or village-level references were excluded. The literature review utilized both peer-reviewed journal articles and a limited selection of gray literature: a literature that is not peer reviewed and indexed by major database but are valuable to research (Government report, NGO publications, etc). The literature was sourced through Google Scholar and research gate using keywords and combinations such as 'Ethnobotany', 'Traditional knowledge', 'Indigenous knowledge', 'Ethnic group', and 'Central Nepal' following Kugley et al. (2017). A total of 102 relevant studies were identified, sorted, and reviewed through a Forward-Backward Reference Searching approach, as Wirtz et al., (2016) recommended. The scientific names and synonyms of plant species mentioned in the reviewed studies were verified through the World Flora Online database (http://www.worldfloraonline.org) to ensure taxonomic accuracy. Data from multiple published sources were compared and the names of plants were checked in standard botanical database to verify data ensuring consistency and reliability.

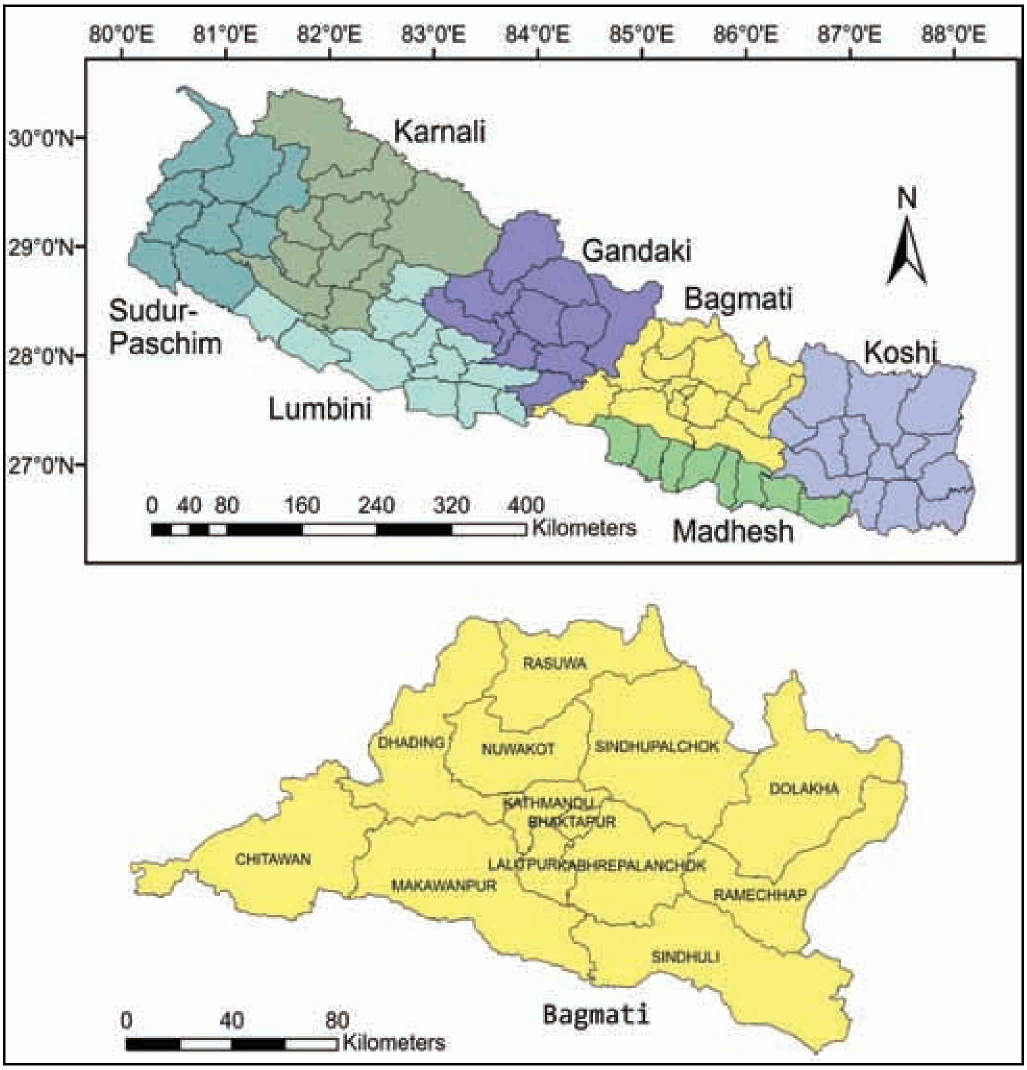

Figure 1: Map of the study sites

Results and Discussions

Plants diversity

About 2245 use reports of 557 plant species belonging to 139 families along 15 use categories were recorded (Appendix 1). The Fabaceae family was documented as the most diverse, with 43 species, followed by Asteraceae with 33 species and Lamiaceae with 27 species. These families were widely distributed across ecosystems, likely due to their adaptability and economic importance. Moderate species counts were observed in families like Poaceae (20 species), Solanaceae (14 species), and Moraceae (14 species), known for their roles in food, medicine, and agriculture. The dominance of key families such as Fabaceae, Asteraceae, and Poaceae in Makawanpur (Bhattarai & Tamang, 2017) aligns with our findings, highlighting their medicinal importance, ecological adaptability, and need for conservation and further study. Families such as Malvaceae, Lauraceae, Rosaceae, and Cucurbitaceae were represented by 11 to 13 species, showing moderate levels of diversification. As the data continued, additional families with fewer species were identified, with some, including Amaryllidaceae and Verbenaceae, containing around five species each, indicating specialized ecological roles. Families like Acoraceae, Actinidiaceae, and Agavaceae are represented by a single species, likely reflecting specific ecological adaptations or restricted distributions. This distribution was used to highlight both the diversity within major plant families and the narrower specialization in others, underscoring the ecological variety across plant taxa.

The ethnobotanical study's finding shows that the richness and depth of traditional knowledge among Nepalese communities, especially medicinal knowledge is the most documented use category which is over 1800. This finding is similar to the Farnsworth (1994), who mentioned the use of plant-based medicine is one of the widely used system of indigenous knowledge.

Futhermore, the findings resonate with those of Balemie & Kebebew (2006), who reported that more than 75% of rural inhibitants in developing nation use traditional medicines because thye lack access to modern medicine. In Nepal, Community knowledge that has been passing down through years is filling up the gap between healthcare and modern medicine in remote areas specially mountaineous region (Zewdu & Demissie, 2001).

Plant use categories

The ethnobotanical data categorized by use, highlights the predominant role of plants in traditional medicine, which is by far the most significant category with 1,822 use reports which is about 81% of the total use report. This dominance indicates the strong reliance of local communities on plants for health and healing purposes, reflecting rich traditional knowledge of medicinal flora. The use of a higher number of reports on medicinal plants (1822) is in agreement with Kunwar et al. (2018) observations from western Nepal and Uprety et al. (2010) study from central Nepal about the importance of traditional medicine in local health-care systems.

Beyond medicinal use, plants are utilized for a variety of purposes, including tonic plants (37 reports), wild edible fruits (44 reports), and vegetables and pickles (78 reports), showcasing their contribution to nutrition and food security.

Other important uses include fuelwood (57 reports) and fodder (62 reports), which play essential roles in daily subsistence, particularly in rural areas. Non-timber forest products (NTFPs) that contribute to the economy account for 41 reports, underscoring the economic importance of plant resources beyond direct consumption. Religious (20 reports), cultural (13 reports), ethnic uses (10 reports), and ritual (6 reports) categories reflect the deep connection between plants and spiritual or cultural practices. Less frequent categories, such as agricultural tools (3 reports) and plants used for oil (10 reports), suggest specific but vital uses in certain communities (Figure 2). All these utilization patterns establish the multi-functional nature of plants that provide for basic human needs, along with cultural practices, within Nepalese communities.

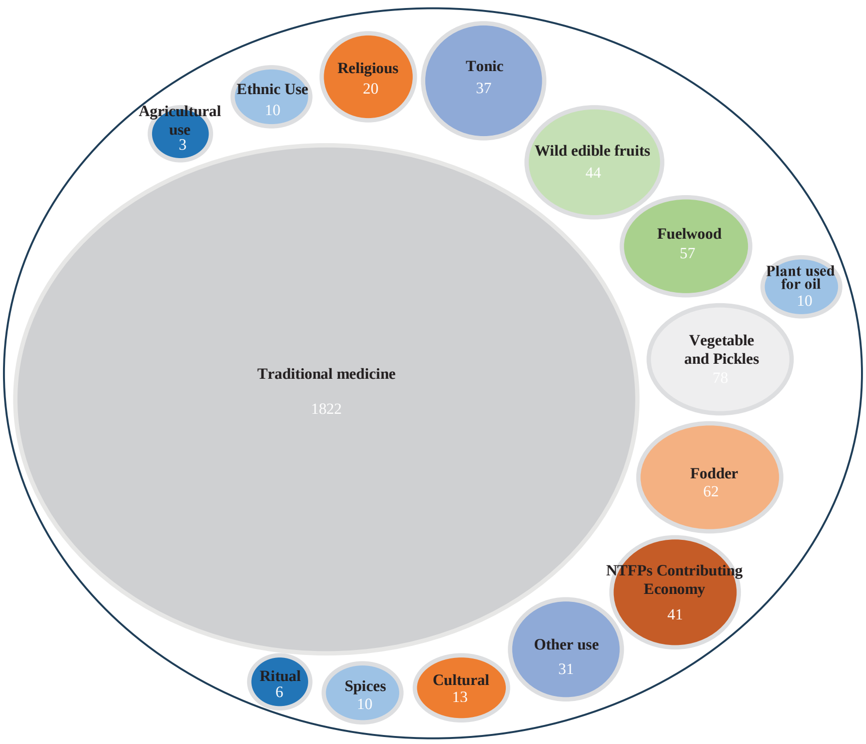

Figure 2: Number of plant species under various use categories

Distribution of ethnobotanical knowledge accordingly to geographical locations

The number of species recorded appears to correlate with the intensity of ethnobotanical studies in each district and the geographical and ecological diversity present. Districts like Makwanpur (270 species), Sindhuli (149 species), and Dolakha (127 species) had higher species records,likely due to more extensive field studies and their diverse habitats that range from subtropical lowlands to mid-hills, offering a variety of ecological niches conducive to plant diversity (Figure 3). Also, a study by Chhetri et al. (2024) shows that Nepal's mid-mountain regions are rich in medicinal and aromatic plants (MAPs), with 819 species reported, many of which are endemic. In contrast, districts such as Sindupalchowk (57 species) and Rasuwa (45 species) had lower species counts, which may be influenced by less ethnobotanical research conducted in these areas, possibly due to their remote locations and challenging terrains. Additionally, variations in the degree of human interaction with the environment, local traditions of plant use, and accessibility for researchers also played a role in the spatial distribution of species records (Kunwar et al., 2022).

The district with easier accessibility and more ethnic groups such as Makawanpur was found with more research papers resulting in a higher number of ethnomedicinal plant records. This suggests that the availability of more publications is from the districts where active ethnobotanical utilization is linked. On the contrary, a lower number of medicinal plants were used at high-altitude districts (Rasuwa, Sindhupalchok) and city-dominated districts (Kathmandu) likely to be attributed to the low human population, high outmigration, rugged physiography, and access to other easy and accessible alternatives as it has been argued that the people living in low-acculturation areas may use fewer plant species for medicine (Vandebroek et al., 2004). This spatial variability in species documentation underscores the importance of comprehensive surveys across all ecological zones. It suggests that areas with lower species records may harbor undocumented biodiversity and that research efforts in such districts may need to be intensified. The overall trend demonstrates that species richness is not only a reflection of ecological diversity but also of the spatial distribution and intensity of ethnobotanical studies conducted across Bagmati province.

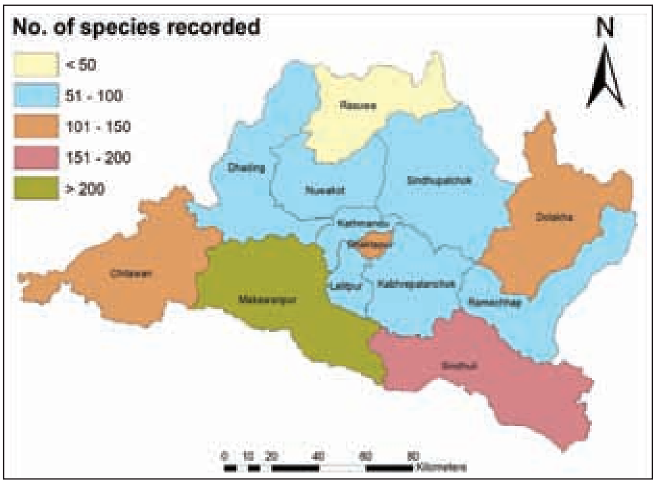

Figure 3: Number of plant species recorded as per districts

Ethnicity and plant utilization

The ethnobotanical study results, categorized by ethnicity, reveal various number of plant species documented for different ethnic groups in Nepal. The Tamang and Newar groups recorded the highest levels of species diversity, with 327 and 257species respectively (Figure 4). Long-standing practices likely influenced this extensive agriculture, medicine, and trade knowledge. Similar patterns were observed among the Brahmin and Chhetri groups, each with 242 species recorded, reflecting their historical roles in religious and agricultural traditions. Between 100 and 201 species were documented by the Danuwar, Bankariya, Jirel, Damai, and Kami communities, indicating substantial engagement with local flora. Intermediate levels of knowledge were noted among the Sherpa, Tharu, Magar, and Majhi groups, with 62 to 95 species being recorded. The Gurung, Pahari, Sarki, and Chepang groups recorded fewer species, with the Chepang documenting only 10. The selection and utilization of knowledge of useful plant resources reflects a strong correlation between ethnicity and ethnobotanical knowledge in Nepal as the traditional people around the world possess unique knowledge of plant resources where larger ethnic communities tend to have a more extensive repertoire of plant species, however, smaller communities may still hold valuable, yet under-documented, traditional knowledge of local plant species (Martin, 1995). This emphasizes the need for more focused ethnobotanical studies to capture the full breadth of knowledge across all ethnic groups. Moreover, the mid-hills and mountainous districts hold the highest number of medicinal plant species in use. It may be linked to the larger human population and ethnic groups of that area possibly due to cultural heterogeneity, and accessibility (Kunwar et al., 2022).

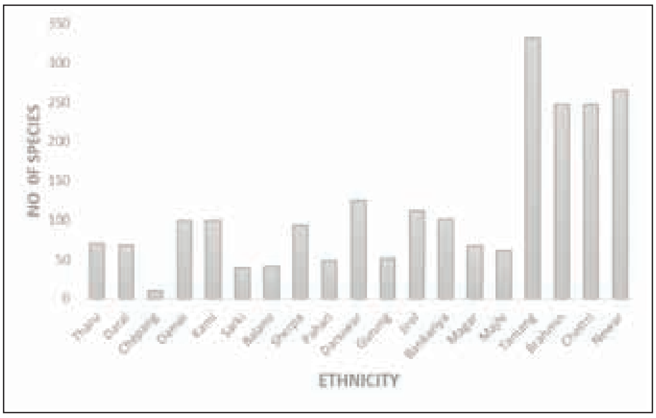

Figure 4: Number of species vs ethnicity

Conclusion

This study documents the traditional use of plant by the indigenious communities residing in the Bagmati Province of Nepal. Despite of the occurance of different ecological zones, people here are connected by living heritage i.e. traditional knowledge. The availability of species according to the abundance of plant family and the use of them in traditional way presents the balance in the ecosystem. The mostly used system is medicinal system, where bark, root, fruit, etc of plants are used. Other uses such as fuel, food, tools, and culturally and traditionally are also equally prevalent in this province.

Future initiatives must include comprehensive surveys in previously unexplored areas, integration of indigenous knowledge into conservation policy, and the establishment of frameworks that protect indigenous knowledge while ensuring equitable benefit-sharing. Bioprospecting, combined with modern science, can be a great achievement in the fields of medicine and agriculture; the knowledge will get scientific validation. Hence, the traditional knowledge will remain accessible in the future. Intergenerational knowledge transmission can be facilitated through education programs that reverse the loss of traditional practices, while scientific verification of medicinal uses can enhance their acceptance. Preservation of ethnobotanical data is critical not only to preserve biodiversity but also to develop pharmaceuticals, ensure sustainable resource management, and conserve culture. It depends on achieving the balance between traditional use and sustainable development- both in respecting cultural heritage and conserving ecological integrity.

Author Contributions

First author and third author did data acquisition, all author did data Analysis. Manuscrpit preparation had done by First author, second author and third author. Second author and fourth author did manuscript review.

Acknowledgements

I would like to express my heartfelt gratitude to Mr. Jagannath Subedi and Mr. Gopalchandra Bhattarai for their assistance in accessing and interpreting relevant gray literature, which significantly contributed to the depth of this review. I am sincerely thankful to my peers; Sharada Poudel and Rewati Acharya, for their meaningful feedback and thoughtful suggestions that helped improve the clarity and structure of the manuscript.

References

Albuquerque, U., & Hanazaki, N. (2009). Five problems in current ethnobotanical research-and some suggestions for strengthening them. *Human Ecology*, *37*, 653-661. https://doi.org/10.1007/s10745-009-9259-9

Bagmati Province Government. (2024). *Second periodic plan of Bagmati Province (FY 2024/25–2028/29)*.

Balami, N. P. (2004). Ethnomedicinal uses of plants among the Newar community of Pharping village of Kathmandu district, Nepal. *Tribhuvan University Journal*, *24*(1), 13-19.

Balemie, K., & Kebebew, F. (2006). Ethnobotanical study of wild edible plants in Derashe and Kucha districts, South Ethiopia. *Journal of Ethnobiology and Ethnomedicine*, *2*, 53. https://doi.org/10.1186/1746-4269-2-53

Baral, S. R., & Kurmi, P. P. (2006). *A compendium of medicinal plants in Nepal*. Mrs. Rachana Sharma.

Bhaila, A., Shakya, S., Kunwar, B., Baral, B., Chaudhary, S., & Munankarmi, N. N. (2022). Ethnomedicinal exploration of plants utilized by the people of Suryabinayak Municipality in

Bhaktapur District, Nepal. *Vegetos, 35*(3), 763-774.

Bhattarai, N. K. (1989). Ethnobotanical studies in Central Nepal: The ceremonial plant-foods. *Contributions to Nepalese Studies, 16*(1), 35-41.

Bhattarai, N. K. (1990). Herbal folk medicines of Kabhrepalanchok district, Central Nepal. *International Journal of Crude Drug Research, 28*(3), 225-231.

Bhattarai, S. B. S., & Tamang, R. T. R. (2017). Medicinal and aromatic plants: A synopsis of Makawanpur district, Central Nepal. *International Journal of Indigenous Herbs and Drugs*, 6-15.

Chaudhary, R. P., Uprety, Y., Devkota, S., Adhikari, S., Rai, S. K., & Joshi, S. P. (2020). Plant biodiversity in Nepal: Status, conservation approaches, and legal instruments under new federal structure BT. In M. Siwakoti, P. K. Jha, S. Rajbhandary, & S. K. Rai (Eds.), *Plant Diversity in Nepal* (pp. 167-206). Botanical Society of Nepal.

Chhetri, R. B., & Gauchan, D. P. (2007). Traditional knowledge on fruit pulp processing of Lapsi in Kavrepalanchowk District of Nepal. *Indian Journal of Traditional Knowledge, 6*(1), 46-49

Chhetri, V. T., Shrestha, S., Thapa, S. B., & Timilsina, S. (2024). The status and role of medicinal and aromatic plants (MAPs) in the Nepalese livelihood. *Authorea*. https://doi.org/10.22541/au.170665962.25962502/v1

Dangol, D., & Gurung, S. (2008). Ethnobotany of the Tharu tribe of Chitwan district, Nepal. *Pharmaceutical Biology, 29*, 203-209. https://doi.org/10.3109/13880209109082879

Dhital, A. P., Paudel, M., Karki, S., Kafle, S., Siwakoti, M., & Lamichhane, D. (2021). Traditional knowledge on use of medicinal plants by Tamang community of Dolakha, Nepal. *Journal of Plant Resources, 19*(1), 192-203.

Dulal, K., Chaudhary, S., Uprety, Y., Shrestha, N., Shakya, S., & Munankarmi, N. (2022). Ethnomedicinal plants used by the local people of Changunarayan Municipality, Bhaktapur, Nepal. *Ethnobotany Research and Applications, 23*, 1-27.

Fabricant, D. S., & Farnsworth, N. R. (2001). The value of plants used in traditional medicine for drug discovery. *Environmental Health Perspectives, 109*(suppl 1), 69-75.

Farnsworth, N. R. (1994). *Ethnobiology and the search for new drugs* (pp. 42-59). John Wiley & Sons Ltd.

Gautam, R. K., & Dhakal, D. P. (2023). Knowledge distribution and ethnobiology in Majhi community of Makawanpur, Nepal. *International Research Journal of MMC (IRJMMC), 4*(1), 49-56.

Giday, M., Asfaw, Z., & Woldu, Z. (2009). Medicinal plants of the Meinit ethnic group of Ethiopia: An ethnobotanical study. *Journal of Ethnopharmacology, 124*(3), 513-521.

Gurung, M. (2023). Ethnomedicinal study of plants used by Newar community in Sindhupalchowk district, Nepal. *Journal of Plant Resources, 21*(1), 32-39.

Gurung, S., & Subedi, S. (2021). Ethnobotanical application of medicinal plants by the Gurung community of Siddhalek rural municipality, Dhading, Nepal. *Asian Journal of Pharmacognosy, 4*(3), 13-21.

Heinrich, M., & Bremner, P. (2006). Ethnobotany and ethnopharmacy - their role for anti-cancer drug development. *Current Drug Targets, 7*(3), 239-245.

Islam, R. (2006). *Role of plant medicine in health care and improving nutritional standard in rural area of Bangladesh*. 1-30.

Islam, R. (2006). Role of plant medicine in health care and improving nutritional standard in rural area of Bangladesh. *National Seminar on Diversity of Medicinal Plants and Their Sustainable Utilization in Health Care and Improving Nutritional Standard in Rural Areas (Kolkata)* (pp. 1-30).

Joshi, K., Joshi, R., & Joshi, A. R. (2011). Indigenous knowledge and uses of medicinal plants in Macchegaun, Nepal. *Indian Journal of Traditional Knowledge, 10*(2), 281-286.

Joshi, N., Ghorbani, A., Siwakoti, M., & Kehlenbeck, K. (2020). Utilization pattern and indigenous knowledge of wild medicinal plants among three ethnic groups in Makawanpur district, Central Nepal. *Journal of Ethnopharmacology*, *262*, 113219.

Kafle, R., & Dani, R. S. (2022). Ethnomedicinal practices of plants in Danuwar community of Dudhauli Municipality, Sindhuli district, Central Nepal. *Journal of Plant Resources*, *20*(2), 211-234.

Karki, S., Dhital, A. P., Uprety, Y., & Ghimire, S. K. (2023). Medicinal plants and their use by an ethnic minority Jirel in Dolakha district, Central Nepal. *Ethnobotany Research and Applications*, *25*, 1-29.

Kugley, S., Wade, A., Thomas, J., Mahood, Q., Jørgensen, A. M. K., Hammerstrøm, K., & Sathe, N. (2017). Searching for studies: A guide to information retrieval for Campbell systematic reviews. *Campbell Systematic Reviews*, *13*(1), 1-73.

Kunwar, R. M. (2006). Dhading district, Central Nepal. *Nepal Journal of Science and Technology*, *7*, 65-69.

Kunwar, R. M., Baral, B., Luintel, S., & al., et. (2022). Ethnomedicinal landscape: Distribution of used medicinal plant species in Nepal. *Journal of Ethnobiology and Ethnomedicine*, *18*(34). https://doi.org/10.1186/s13002-022-00531-x

Kunwar, R. M., & Bussmann, R. W. (2008). Ethnobotany in the Nepal Himalaya. *Journal of Ethnobiology and Ethnomedicine*, *4*, 1-8.

Kunwar, R. M., Fadiman, M., Cameron, M., Bussmann, R. W., Thapa-Magar, K. B., Rimal, B., & Sapkota, P. (2018). Cross-cultural comparison of plant use knowledge in Baitadi and Darchula districts, Nepal Himalaya. *Journal of Ethnobiology and Ethnomedicine*, *14*, 1-17.

Kunwar, R. M., Nepal, B. K., Kshhetri, H. B., Rai, S. K., & Bussmann, R. W. (2006). Ethnomedicine in Himalaya: A case study from Dolpa, Humla, Jumla and Mustang districts of Nepal. *Journal of Ethnobiology and Ethnomedicine*, *2*, 1-6.

Lesnikowski, A. C., Ford, J. D., Berrang-Ford, L., Barrera, M., Berry, P., Henderson, J., & Heymann, S. J. (2013). National-level factors affecting planned, public adaptation to health impacts of climate change. *Global Environmental Change*, *23*(5), 1153-1163.

Luitel, D. R., Rokaya, M. B., Timsina, B., & Münzbergová, Z. (2014). Medicinal plants used by the Tamang community in the Makawanpur district of central Nepal. *Journal of Ethnobiology and Ethnomedicine*, *10*, 1-11.

Malla, B., & Chhetri, R. B. (2009). Indigenous knowledge on ethnobotanical plants of Kavrepalanchowk district. *Kathmandu University Journal of Science, Engineering and Technology*, *5*(2), 96-109.

Manandhar, N. P. (1991). Medicinal plant-lore of Tamang tribe of Kabhrepalanchok district, Nepal. *Economic Botany*, *45*(1), 58-71.

Martin, G. J. (1995). *Ethnobotany: A methods manual*. Chapmany Hall.

Moktan, D. B. (2023). Indigenous knowledge on health and use of herbal plants as domestic: A case study of illiterate an indigenous caste 'Bankariya.' *Southwestern Research Journal (SRJ)*.

Nagarkoti, S., & Shrestha, S. J. (2022). Ethnomedicinal plants used by Pahari community of Shikharpa village, Lalitpur, Nepal. *Journal of Plant Resources*, *20*(1), 159-172.

National Statistics Office (2023). *National population and housing census 2021 (National report)*. Government of Nepal.

Nepali, K. B., Pathak, M. L., Shah, S., & Lamichhane, D. (2020). Documentation of indigenous knowledge on plants used by Tamang community of Nuwakot district, Central Nepal. *Journal of Plant Resources*, *18*(1), 252.

Panta, A. (2019). *Medico-ethnobiology, indigenous technology and indigenous knowledge system of Newar ethnic group in Khokana village of Lalitpur District, Nepal*. Central Department of Zoology.

Poudel, M., & Singh, N. B. (2016). Medical ethnobiology and indigenous knowledge system found in Darai ethnic group of Chitwan, Nepal. *Journal of Institute of Science and Technology*, *21*(1), 103-111.

Pradhan, S. P., Chaudhary, R. P., Sigdel, S., & Pandey, B. P. (2020). Ethnobotanical knowledge of Khandadevi and Gokulganga rural municipality of Ramechhap district of Nepal. *Ethnobotany Research and Applications*, *20*, 1-32.

Pratik, A., Roshan, K. C., Kayastha, D., Thapa, D., Shrestha, R., Shrestha, T. M., & Gyawali, R. (2011). Phytochemical screening and anti-microbial properties of medicinal plants of Dhunkharka community, Kavrepalanchowk, Nepal. *International Journal of Pharmacy and Biological Archives*, *2*, 1663-1667.

Rai, S. K., Subedi, S., & Mishra, S. (2004). Utilization pattern of medicinal plants in Thumpakhar, Village Development Committee, Sindhupalchok, Nepal. *Botanica Orientalis*, *4*(1), 75-78.

Rijal, A. (2008). Living knowledge of the healing plants: Ethno-phytotherapy in the Chepang communities from the Mid-Hills of Nepal. *Journal of Ethnobiology and Ethnomedicine*, *4*, 1-10.

Rokaya, M. B., Münzbergová, Z., & Timsina, B. (2010). Ethnobotanical study of medicinal plants from the Humla district of Western Nepal. *Journal of Ethnopharmacology*, *130*(3), 485–504.

Shah, D. P., & Singh, N. B. (2014). Ethnobiological study of the Majhi of Sindhuli district. *Nepalese Journal of Zoology*, *2*(1), 44-50.

Shah, S., & Lamichhane, D. (2017). Documentation of indigenous knowledge on plants used by Tamang community of Kavrepalanchok district, Central Nepal. *Journal of Plant Resources*, *15*, 45-51.

Sharma, L. N., & Bastakoti, R. (2010). Ethnobotany of Dioscorea L. with emphasis on food value in Chepang communities in Dhading district, central Nepal. *Botanica Orientalis: Journal of Plant Science*, *6*.

Shrestha, N., Prasai, D., Shrestha, K. K., Shrestha, S., & Zhang, X.-C. (2014). Ethnomedicinal practices in the highlands of Central Nepal: A case study of Syaphru and Langtang village in Rasuwa district. *Journal of Ethnopharmacology*, *155*(2), 1204-1213. https://doi.org/10.1016/j.jep.2014.07.002

Shrestha, P. M., & Dhillion, S. S. (2003). Medicinal plant diversity and use in the highlands of Dolakha district, Nepal. *Journal of Ethnopharmacology*, *86*, 81-96.

Tamang, G. (2003). An ethnobiological study of the Tamang people. *Our Nature*, *1*(1), 37-41.

Thapa, S. (2012). Medico-ethnobotany of Magar community in Salija VDC of Parbat district, Central Nepal. *Our Nature*, *10*(1), 47-53.

Timilsina, S. H., & Singh, N. B. (2015). Ethnobiology and indigenous knowledge about medicinal animals and plants in the Balami ethnic group in Nepal. *Journal of Institute of Science and Technology*, *19*(2), 79. https://doi.org/10.3126/jist.v19i2.13857

Uprety, Y., Asselin, H., Boon, E. K., Yadav, S., & Shrestha, K. K. (2010). Indigenous use and bio-efficacy of medicinal plants in the Rasuwa district, Central Nepal. *Journal of Ethnobiology and Ethnomedicine*, *6*(3). https://doi.org/10.1186/1746-4269-6-3

Vandebroek, I., Van Damme, P., Van Puyvelde, L., Arrazola, S., & De Kimpe, N. (2004). A comparison of traditional healers' medicinal plant knowledge in the Bolivian Andes and Amazon. *Social Science & Medicine*, *59*(4), 837-849.

Wirtz, B., Pistoia, A., Ullrich, S., & Göttel, V. (2016). Business models: Origin, development and future research perspectives. *Long Range Planning*, *49*. https://doi.org/10.1016/j.lrp.2015.04.001

Wyk, B. V, Oudtshoorn, B. V, & Gericke, N. (1997). *Medicinal Plants of South Africa*. Briza Publication.

Zewdu, M., & Demissie, A. (2001). *Conservation and sustainable use of medicinal plants in Ethiopia*. Institute of Biodiversity Conservation and Research.

Appendix 1: List of plant species with their family, reported districts and uses

S. N.	Species/Family	Reported districts	Uses
1	*Abelmoschus esculentus* (L.) Moench Malvaceae	Kavrepalanchowk	Juice for cuts, wounds, urinary problems, abdominal disorder, constipation, diarrhea, general debility; tender fruits as vegetable (Malla & Chhetri, 2009)
2	*Abelmoschus moschatus* moench Malvaceae	Chitwan	Promote healing of cuts and wounds (Dangol & Gurung, 2008)
3	*Abrus precatorius* L. Fabaceae	Sinduli, Nuwakot, Bhaktapur, Makawanpur, Sindupalchowk	Eye infections (Shah & Singh, 2014), cleaning dust and foreign particles, eye impurities, itching
4	*Abutilon indicum* (L.) Sweet Malvaceae	Chitwan	Extract pus (Dangol & Gurung, 2008)
5	*Acacia catechu* (L.f.) Willd. Fabaceae	Makawanpur, Sinduli, Ramechaap	Cough ,Toothache (Shah & Singh, 2014), Used in cure of body inflammation, blood purifier, chest pain, asthma, cold, skin diseases, fever and as timber and fodder (Bhattarai & Tamang,2017; Joshi et al., 2020; Pradhan et al., 2020; Moktan,2023)
6	*Acacia nilotica* (L.) Willd. ex Delile Fabaceae	Chitwan, Makawanpur	Dysentery, diarrhea in children; flowers soaked in water used to quench thirst (Bhattarai & Tamang, 2017; Dangol & Gurung, 2008)
7	*Acer oblongum* Wall. ex DC. Aceraceae	Makawanpur	Leaves used as fodder (Moktan, 2023)
8	*Achyranthes aspera* L. Amaranthaceae	Makawanpur, Chitwan, Bhaktapur, Lalitpur, Sinduli, Kathmandu	Fever, sinusitis, toothache, itching, headaches, stop bleeding, placenta issues, stomach pain, typhoid; used as toothbrush (Balami, 2004; Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022)
9	*Achyranthes bidentata* Blume Amaranthaceae	Chitwan, Makawanpur, Sinduli, Dolakha, Nuwakot, Kavrepalanchowk, Lalitpur	Toothache, urinary problems, clean teeth, increase body temperature, childbirth, asthma, respiration problems, typhoid, dysentery, fever, pregnancy; for expelling placenta in animals; stem as toothbrush (Malla & Chhetri , 2009; Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Joshi et al. 2020;Nagarkoti & Shrestha, 2022; Karki et al., 2023)
10	*Acmella oleracea* (L.) R.K.Jansen Asteraceae	Bhaktapur	Toothache (Bhaila et al., 2022)
11	*Aconitum ferox* Wall. Ranunculaceae	Ramechaap, Makawanpur, Kathmandu	Root paste/powder for fever, diarrhea, vomiting, stomach problems, indigestion; roots powder mixed with food to kill rodents (Balami, 2004; Bhattarai & Tamang , 2017; Pradhan et al., 2020)
12	*Aconitum lethale* Griff. Ranunculaceae	Rasuwa, Makawanpur	Tuber used for fever (detoxified using milk or cow urine before use) (Shrestha et al., 2014; Joshi et al., 2020)
13	*Acorus calamus* L. Acoraceae	Nuwakot, Sinduli, Chitwan, Makawanpr, Dhading, Lalitpur, Rasuwa, Dolakha, Bhaktapur, Sindupalchowk	Cough, bronchitis, neck problem, kidney diseases, insomnia, diarrhea, antidote, tonsillitis, asphyxia, anti-lice, sore throat, respiratory problems, cold, fever, fracture (Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022;Gautam & Dhakal, 2023; Gurung, 2023; Karki et al., 2023; Moktan, 2023)
14	*Adiantum caudatum* L. Pteridaceae	Chitwan	Tied around wrist for nightly fever, cooling effect, athlete's foot, wounds (Dangol & Gurung, 2008)
15	*Adina cordifolia* (Roxb.) Brandis Rubiaceae	Chitwan, Sinduli	Cooling effect, athlete's foot, wounds (Dangol & Gurung, 2008; Kafle & Dani, 2022)
16	*Aechmanthera gossypina* (Wall.) Nees Acanthaceae	Makawanpur	Diarrhea (Joshi et al., 2020)
17	*Aegle marmelos* (L.) Corrêa Rutaceae	Sinduli, Chitwan, Ramechaap, Makawanpur, Dhading, Bhaktapur	Diarrhea, constipation, diabetes, dysentery, gastritis, typhoid, jaundice, inflammation, toothache, stomach issues, body strength, fever/cooling, bleeding nose; fruits edible; leaves for religious worship; fodder (Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Moktan, 2023)
18	*Aeschynomene aspera* L. Fabaceae	Makawanpur	Plant paste applied to forehead for headache caused by sinusitis (Moktan, 2023)
19	*Aerva lanata* (L.) Juss. Amaranthaceae	Chitwan	Juice with leaf buds of Karma and Gurench vine given for fever (Dangol & Gurung, 2008)
20	*Agave americana* L. Asparagaceae	Bhaktapur, Makawanpur	Constipation (Bhattarai & Tamang, 2017; Dulal et al., 2022)
21	*Ageratina adenophora* (Spreng.) R.M.King & H.Rob. Asteraceae	Lalitpur, Dolakha, Nuwakot, Bhaktapur, Sinduli, Makawanpur Kavrepalanchowk, Sindupalchowk	Cuts, wounds, fever, boils, eye insomnia; green manure, bio-briquette, fodder; juice for cough, cold, sinusitis (Rai et al., 2004; Malla & Chhetri, 2009; Shah & Singh, 2014; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Nepali et al., 2020; Dhital et al., 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023; Moktan, 2023)
22	*Ageratum conyzoides* L. Asteraceae	Bhaktapur, Chitwan, Makawanpur	Blood clotting, bone fracture, cuts and wounds, setting dislocated bones, stopping bleeding (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Joshi et al., 2020; Bhaila et al., 2022; Moktan, 2023)
23	*Albizia julibrissin* Durazz. Fabaceae	Nuwakot, Makawanpur	Firewood, banana ripening (Bhattarai & Tamang, 2017; Nepali et al., 2020)
24	*Allium hypsistum* Stearn Amaryllidaceae	Nuwakot, Lalitpur, Dhading	Cough, cold, flu, stomach pain, muscle ache, scabies, corneal ulcer; used as spice (Timilsina & Singh, 2015; Panta, 2019; Gurung & Subedi, 2021)
25	*Allium sativum* L. Amaryllidaceae	Chitwan, Makawanpur, Kavrepalanchowk, Sinduli, Dolakha, Bhaktapur	Tonsillitis, fever, scorpion bite, earache, gastric issues, constipation, cough, cold, ringworm; used as spice and stimulant; roasted tubers eaten fresh/pickled; leaves as vegetables; oil for skin rashes (Malla & Chhetri, 2009; Poudel & Singh, 2016; Dhital et al., 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023)
26	*Allium wallichii* Kunth Amaryllidaceae	Dolakha, Makawanpur, Kathmandu	Cough and cold (Balami, 2004; Joshi et al., 2020; Dhital et al., 2021)
27	*Alnus nepalensis* D.Don Betulaceae	Kavrepalanchowk, Nuwakot, Makawanpur	Diarrhea, dysentery, burns, cuts, wounds; wood for furniture, construction, fuel; fodder (Malla & Chhetri, 2009; Joshi et al., 2020; Nepali et al., 2020)
28	*Aloe vera* (L.) Burm.f. Asphodelaceae	Sinduli, Nuwakot, Chitwan, Makawanpur, Bhaktapur, Dhading, Lalitpur, Sindupalchowk	Burns, burnt wounds, menstruation regulation, intestinal wounds, jaundice, skin allergies, high blood pressure, body temperature, kidney stones, weakness, skin infection, intestinal worms (Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Dhital et al., 2021; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Gurung, 2023; Karki et al., 2023)
29	*Alsophila spinulosa* (Hook.) R.M.Tryon Cyatheaceae	Dolakha	Rhizome paste to expel spines from body (Karki et al., 2023)
30	*Alstonia scholaris* (L.) R.Br. Apocynaceae	Makawanpur, Sinduli	Wood for musical instrument (madal); antihelminthic, anticholera, diabetes, epilepsy, sterility, abortion, weakness (Bhattarai & Tamang, 2017; Joshi et al., 2020; Kafle & Dani, 2022; Moktan, 2023)
31	*Alternanthera sessilis* (L.) DC. Amaranthaceae	Sinduli, Makawanpur	Muscle ache, scabies, corneal ulcer (Shah & Singh, 2014; Joshi et al., 2020; Kafle & Dani, 2022)
32	*Amaranthus spinosus* L. Amaranthaceae	Chitwan, Bhaktapur, Sinduli, Makawanpur, Kathmandu	Lower fever, skin problems, labor pain reducer (Balami, 2004; Dangol & Gurung, 2008; Poudel & Singh, 2016; Joshi et al., 2020; Kafle & Dani, 2022)

S. N.	Species/Family	Reported districts	Uses
33	*Amaranthus viridis* L. Amaranthaceae	Lalitpur	Leaf juice/curry for constipation relief (Nagarkoti & Shrestha, 2022)
34	*Amomum subulatum* Roxb. Zingiberaceae	Bhaktapur, Dhading, Makawanpur	Prevent sore throat, cold Bhattarai & Tamang, 2017; Gurung & Subedi, 2021; Dulal et al., 2022)
35	*Amorphophallus paeoniifolius* (Dennst.) *Nicolson* Araceae	Sinduli	Swollen body (Kafle & Dani, 2022)
36	*Ampelocissus divaricata* (Wall. ex M.A.Lawson) Planch Vitaceae	Makawanpur	Eyesight improvement (Joshi et al., 2020)
37	*Ananas comosus* (L.) Merr. Bromeliaceae	Chitwan, Sinduli	Heat, typhoid (Bhattarai & Tamang, 2017; Gurung & Subedi, 2021; Dulal et al., 2022)
38	*Anaphalis busua* DC. Asteraceae	Makawanpur	Cuts and wounds (Joshi et al., 2020)
39	*Anaphalis triplinervis* Sims ex C.B.Clarke Asteraceae	Makawanpur, Kathmandu	Cuts and wounds (Balami, 2004; Joshi et al., 2020)
40	*Anethum graveolens* L. Apiaceae	Chitwan, Sinduli	Lactation enhancer, tonic for pregnant women (Poudel & Singh, 2016; Kafle & Dani, 2022)
41	*Anisomeles indica* (L.) Kuntze Lamiaceae	Makawanpur	Tonic (Joshi et al., 2020)
42	*Anthogonium gracile* Wall. ex Lindl. Orchidaceae	Dolakha	Pseudobulb chewed to smoothen the neck, neck pain relief (Karki et al., 2023)
43	*Areca catechu* L. Arecaceae	Chitwan, Lalitpur, Sinduli	Scars, eczema, scar removal (Poudel & Singh, 2016; Panta, 2019; Kafle & Dani, 2022)
44	*Argemone mexicana* L. Papaveraceae	Ramechaap, Sinduli	Skin disease, jaundice, cough, cold, eye troubles (Pradhan et al., 2020; Kafle & Dani, 2022)
45	*Argentina festiva* (Soják) Soják Rosaceae	Makawanpur, Nuwakot	Fever, headache, toothache (Joshi et al., 2020; Nepali et al., 2020)
46	*Argentina lineata* (Trevir.) Soják Rosaceae	Dolakha	Stomach disorder, gastritis, worms, tonsillitis (Dhital et al., 2021; Karki et al., 2023)
47	*Artemisia indica* Willd. Asteraceae	Chitwan, Sindupalchowk, Makawanpur, Lalitpur, Dolakha, Rasuwa, Nuwakot, Kathmandu	Used in baby naming function, child bath, wounds, poison, intestinal worms, heating body, dislocated bones, mosquito repellent, insecticide, body odor, rheumatic disease, incense, cough, religious value, scabies, ringworm, diabetes (Balami, 2004; Rai et al., 2004; Bhattarai & Tamang, 2017; Poudel & Singh, 2016; Joshi et al., 2020; Nagarkoti & Shrestha, 2022; Karki et al., 2023; Moktan, 2023)
48	*Artemisia vulgaris* L. Asteraceae	Chitwan, Makawanpur, Nuwakot, Dhading, Lalitpur, Bhaktapur, Sinduli	Cough, asthma, cuts, wounds, scabies, bleeding, skin disease, stomach pain, gastritis, anti-leech, uterine prolapse, sprain, cold, pressure reduction, sugar decrease, antihelminthic, typhoid, allergy, repellent for bugs and flies (Dangol & Gurung, 2008; Shah & Singh, 2014; Timilsina & Singh, 2015; Panta, 2019; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023)
49	*Artocarpus heterophyllus* Lam. Moraceae	Kavrepalanchowk, Makawanpur	Skin disease, diarrhea, dysentery, glandular swelling, abscesses; fruits eaten fresh/as vegetable/pickled; seeds roasted; leaves as fodder (Malla & Chhetri, 2009; Moktan, 2023)
50	*Artocarpus lacucha* Roxb. ex Buch.-Ham. Moraceae	Makawanpur, Sinduli, Chitwan	Mumps, diarrhea, wounds, boils, gastritis, indigestion; ripe fruits eaten fresh; young shoots as vegetable; leaves as fodder (Poudel & Singh, 2016; Kafle & Dani, 2022; Moktan, 2023)
51	*Asparagus racemosus* Willd. Asparagaceae	Makawanpur, Sinduli, Bhaktapur, Lalitpur, Dolakha, Rasuwa, Kathmandu	Paralysis, tonic, nerve disease, body pain, lactation, dysentery, cough, cold, fever, inflammation, expelling animal placenta, urine blockage, venereal diseases; tender shoots eaten as vegetable/pickle (Balami, 2004; Luitel et al., 2014; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Nagarkoti & Joshi et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Karki et al., 2023; Moktan, 2023)
52	*Astilbe rivularis* Buch.-Ham. Saxifragaceae	Ramechaap, Makawanpur, Dolakha, Rasuwa, Lalitpur, Kathmandu	Pain killer, muscular swelling, gastritis, indigestion, delivery stomach issues, tonic for pre/post pregnancy, fracture, body pain, menstruation problems, over-bleeding, cough (Balami, 2004; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
53	*Avena sativa* L. Poaceae	Ramechaap	Spikes as laxative, grain powder for indigestion; grains edible and used in Hindu worship; plants as fodder (Pradhan et al., 2020)
54	*Azadirachta indica* A.Juss. Meliaceae	Nuwakot, Makawanpur, Chitwan, Lalitpur, Bhaktapur, Ramechaap, Sinduli, Dhading,	Blisters, allergy, malaria, respiratory problems, high blood pressure, fever, toothache, antihelminthic, body pain, pneumonia, dysentery, cough, cold, urinary complaints, sugar control, wounds, worms, skin disease; used as toothbrush (Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023)
55	*Azanza lampas* Alef. Malvaceae	Makawanpur	Fever, stomach issues (Joshi et al., 2020; Moktan, 2023)
56	*Bambusa bambos* (L.) Voss Poaceae	Sinduli	Toothbrush (Kafle & Dani, 2022)
57	*Barleria cristata* L. Acanthaceae	Chitwan, Makawanpur, Kavrepalanchowk	Anemia, throat trouble, indigestion, inflammations, fever, bronchitis, asthma, boils, pimples (Malla & Chhetri, 2009; Poudel & Singh, 2016; Joshi et al., 2020; Moktan, 2023)
58	*Bauhinia purpurea* L. Fabaceae	Makawanpur, Nuwakot, Sindupalchowk, Kathmandu	Fodder; flowers and young shoots cooked as vegetable; skin diseases (Balami, 2004; Rai et al., 2004; Bhattarai & Tamang, 2017; Nepali et al., 2020; Moktan, 2023)
59	*Bauhinia variegata* L. Fabaceae	Makawanpur, Dhading, Ramechaap, Nuwakot, Sindupalchowk, Bhaktapur, Kathmandu	Antivenin, back pain, diarrhea, blood pressure, dysentery, stomach ache, body ache, indigestion; flowers pickled; young shoots/leaves as vegetable; bark/flower juice for bleeding, diabetes; heartwood for construction (Balami, 2004; Rai et al., 2004; Joshi et al., 2020; Nepali et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Moktan, 2023)
60	*Begonia picta* Sm. Begoniaceae	Makawanpur	Wounds, anti-leech (Joshi et al., 2020)
61	*Benincasa hispida* Cogn. Cucurbitaceae	Makawanpur, Dhading,	Coolant; fruits used as vegetable and pickled (Gurung & Subedi, 2021; Moktan, 2023)
62	*Berberis aristata* DC. Berberidaceae	Rasuwa, Makawanpur, Nuwakot, Bhaktapur, Sindupalchowk, Kathmandu	Root bark decoction for diarrhea, jaundice, blood purification; ripe fruits eaten; eye problems, skin problems, eye boils, skin disease (Balami, 2004; Shrestha et al., 2014; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Joshi et al., 2020; Dulal et al., 2022; Gurung, 2023)
63	*Berberis napaulensis* (DC.) Spreng. Berberidaceae	Makawanpur, Dolakha, Lalitpur, Rasuwa, Bhaktapur	Leaf juice for mouth ulcer, throat pain, tonsil, fever; fruit/bark infusion for high blood pressure; bark juice for eye irritation/inflammation (Shrestha et al., 2014; Joshi et al., 2020; Dhital et al., 2021; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
64	*Bergenia ciliata* (Haw.) Sternb. Saxifragaceae	Ramechaap, Makawanpur, Dolakha, Rasuwa, Bhaktapur, Sindupalchowk, Lalitpur, Kavrepalanchowk,	Juice for urinary troubles, menstruation problems, gallstones, kidney stones; tonic, blood purifier, toxin removal, aphrodisiac, diarrhea, fever, vomiting, piles, tumors, heart diseases, asthma, lung problems; rhizome powder for cough, dysentery; flowers boiled/pickled; stem paste for back/joint pain, roundworm infection (Balami, 2004; Rai et al., 2004; Malla & Chhetri, 2009; Shrestha et al., 2014; Bhattarai &

S. N.	Species/Family	Reported districts	Uses
		Kathmandu	Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Bhaila et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
65	*Betula alnoides* Buch.-Ham. ex D.Don Betulaceae	Kavrepalanchowk, Rasuwa, Nuwakot, Makawanpur	Bark boiled and applied to dislocated bone/injury; chewed for sore throat and excessive menstruation; wood for furniture, construction, fuel (Malla & Chhetri, 2009; Shrestha et al., 2014; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017)
66	*Bidens pilosa* L. Asteraceae	Kavrepalanchowk, Makawanpur	Plant juice for cuts and wounds; leaves as fodder; tender shoots cooked as vegetables (Malla & Chhetri, 2009; Moktan, 2023)
67	*Biophytum sensitivum* (L.) DC. Oxalidaceae	Chitwan	Plant juice for fever (Dangol & Gurung, 2008)
68	*Bixa orellana* L. Bixaceae	Chitwan	Fracture (Poudel & Singh, 2016)
69	*Boehmeria platyphylla* D.Don Urticacae	Kavrepalanchowk	Plant decoction for livestock diarrhea/dysentery; root paste for cattle wounds/cuts; leaf juice for fresh cuts/wounds; bark yields threads; leaves as fodder (Malla & Chettri , 2009)
70	*Bombax ceiba* L. Malvaceae	Chitwan, Sinduli, Dhading, Lalitpur, Makawanpur	Flower paste for diarrhea, dysentery, stomach troubles; spine paste for measles; cough, bronchitis, dandruff, micturition disorder (Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Gurung & Subedi, 2021; Nagarkoti & Shrestha, 2022; Moktan, 2023)
71	*Bonnaya ciliata* (Colsm.) Spreng. Scrophulariaceae	Chitwan	Plant paste applied on cuts and wounds (Dangol & Gurung, 2008)
72	*Brassica campestris* L. Brassicaceae	Kavrepalanchowk, Dhading	Back pain, diarrhea; seeds for pickle and oil; seed oil cake for cattle; tender shoots/leaves as vegetables; husks for plastering houses (Malla & Chhetri, 2009; Gurung & Subedi, 2021)
73	*Brassica juncea* (L.) Czern. Brassicaceae	Sinduli	Skin allergy (blisters/rashes) (Kafle & Dani, 2022)
74	*Brassica napus* L. Brassicaceae	Kavrepalanchowk	Seed oil for earaches and cooking; husks for plastering houses/baskets; tender green leaves as vegetable (Malla & Chettri , 2009)
75	*Brassica oleracea* var. botrytis Brassicaceae	Sinduli	Diarrhea, fever, bacterial infection (Shah & Singh, 2014)
76	*Brassica oleracea* L. Brassicaceae	Sinduli	Asthma, fever, cough, rheumatism, stomach disorder; mature leaves fermented for 'gundruk' (Shah & Singh, 2014)
77	*Brassica rapa* L. Brassicaceae	Lalitpur	Seed oil for earaches, muscle/joint pain, bone dislocation, massaging newborns and mothers; mixed with Trachyspermum ammi/garlic for cold, cough; mixed with salt for toothache (Nagarkoti & Shrestha, 2022)
78	*Bridelia retusa* (L.) A.Juss. Euphorbiaceae	Kavrepalanchowk	Bark juice for diarrhea, dysentery, peptic ulcer; ripe fruits eaten fresh; leaves as fodder (Malla & Chettri , 2009)
79	*Brucea javanica* (L.) Merr. Simaroubaceae	Dolakha	Fruits eaten for diarrhea and dysentery (Karki et al., 2023)
80	*Bryophyllum pinnatum* (Lam.) Oken Crassulaceae	Lalitpur, Bhaktapur, Makawanpur	Ear problems, kidney stones, urinary disorders, wounds, piles (Bhattarai & Tamang, 2017; Panta, 2019; Dulal et al., 2022)
81	*Buddleja asiatica* Lour. Scrophulariaceae	Dolakha, Sindupalchowk	Pneumonia, skin diseases (Rai et al., 2004; Dhital et al., 2021)
82	*Butea monosperma* (Lam.) Kuntze Fabaceae	Bhaktapur, Sinduli, Makawanpur	Antihelminthic, intestinal worms, blood dysentery (Bhattarai & Tamang, 2017; Bhaila et al., 2022; Kafle & Dani, 2022)
83	*Cajanus cajan* (L.) Millsp. Fabaceae	Kavrepalanchowk	Young leaves for tongue boils, jaundice, cough, cold, diarrhea, wounds; dry leaves as cattle fodder; seeds for soup and 'daal' (Malla & Chettri , 2009)
84	*Callicarpa macrophylla* Vahl Verbenaceae	Dhading, Chitwan, Makawanpur	Fever, sore throat and tongue, earache (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Gurung & Subedi, 2021)
85	*Calotropis gigantea* (L.) W.T.Aiton Apocynaceae	Bhaktapur, Sinduli Chitwan, Makawanpur	Body pain, thorn removal, boils, foot/sole cracks, muscle ache, fracture, sprain, paralysis; milk juice applied on boils (Dangol & Gurung, 2008; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Bhaila et al., 2022; Kafle & Dani, 2022)
86	*Cannabis sativa* L. Cannabaceae	Lalitpur, Sinduli, Chitwan, Makawanpur, Dolakha, Sindupalchowk, Bhaktapur, Kathmandu	Aerial parts as stimulant, digestive, body warming agent, appetizer; twigs for animal diarrhea; for cold, pain relief, abdominal disorder, constipation, insomnia, insanity (Balami, 2004; Rai et al., 2004; Dangol & Gurung, 2008; Joshi et al., 2020; Luitel et al., 2014; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023; Karki et al., 2023)
87	*Cantharellus odoratus* Cantharellaceae	Sinduli	Stomach disorder, diet (Kafle & Dani, 2022)
88	*Capsicum annuum* L. Solanaceae	Sinduli, Lalitpur, Makawanur	Fruits in pickle for gastric problems, anti-cancer; green fruits pickled/eaten fresh; root for fruit-caused fever; headache (Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Moktan, 2023)
89	*Capsicum frutescens* L. Solanaceae	Sinduli	Swollen stomach (Kafle & Dani, 2022)
90	*Capsicum microcarpum* Cav. Solanaceae	Ramechaap, Bhaktapur	Appetizer, cough; green pods/pickles for indigestion, stomach problems (Pradhan et al., 2020; Dulal et al., 2022)
91	*Carica papaya* L. Caricaceae	Dhading, Sinduli	Fungal infection, jaundice, dengue (Gurung & Subedi, 2021; Kafle & Dani, 2022)
92	*Cassia fistula* L. Fabaceae	Chitwan, Makawanpur, Sinduli, Dhading, Ramechaap, Nuwakot,	Bark for throat problems, cough, cold; fruits for urinary tract infections, diarrhea, constipation, urine retention, rheumatism, burning sensation, snake bite, headache (Dangol & Gurung, 2008; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Kafle & Dani, 2022; Gautam & Dhakal, 2023)
93	*Cassia tora* L. Fabaceae	Dhading, Makawanpur	Cough; plant paste for fever (Gurung & Subedi, 2021; Moktan, 2023)
94	*Celosia argentea* L. Amaranthaceae	Sinduli	Stomach disorder, Diet (Kafle & Dani, 2022)
95	*Centella asiatica* (L.) Urb. Apiaceae	Sinduli, Chitwan, Dhading, Dolakha, Lalitpur, Sindupalchowk, Makawanpur, Bhaktapur, Nuwakot, Kathmandu	Jaundice, High blood pressure, Blood purification, Memory enhancement, Nose bleeding, Blood dysentery, Urination issues, Fever, Uric acid, Leprosy, Swollen limbs, Common cold, Throat pain, Tonsillitis, Pneumonia, Urinary disorder, Coolant (Balami, 2004; Dangol & Gurung, 2008; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Dhital et al., 2021; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Karki et al., 2023)
96	*Hemionitis farinosa* (Forssk.) Christenh. Pteridaceae	Ramechaap	Antihelmintic, Eye sight, Back pain, Joint pain (Pradhan et al., 2020)
97	*Chenopodium album* L. Amaranthaceae	Chitwan, Dhading, Makawanpur	Gastritis, Indigestion (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Joshi et al., 2020; Gurung & Subedi, 2021)
98	*Chenopodium murale* L. Amaranthaceae	Chitwan	Tonic, Joint pain, Body strength (Dangol & Gurung, 2008)
99	*Chlorophytum arundinaceum* Baker Asparagaceae	Makawanpur	Hydrocele, Sterilization, High blood pressure, Joint pain, Body ache (Bhattarai & Tamang, 2017; Joshi et al., 2020)

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100	*Chlorophytum nepalense* (Lindl.) Baker Asparagaceae	Makawanpur, Dolakha, Chitwan	Cold, Cough, Diarrhea, Pickle (fruits), Wound treatment, Skin allergies, Skin rashes (Dangol & Gurung, 2008; Joshi et al., 2020; Karki et al., 2023)
101	*Choerospondias* axillaris (Roxb.) B.L.Burtt & A.W.Hill Anacardiaceae	Dolakha, Lalitpur, Makawanpur, Nuwakot	Cut, Wound (Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Nepali et al., 2020; Karki et al., 2023)
102	*Chromolaena odorata* (L.) R.M.King & H.Rob. Asteraceae	Makawanpur, Sinduli	Fever, Mouth sore, Tonsil, Blood pressure, Diabetes, Ringworm, Indigestion, Rashes, Ornamental (Bhattarai & Tamang, 2017; Joshi et al., 2020; Kafle & Dani, 2022)
103	*Chrysojasminum humile* (L.) Banfi Oleaceae	Kavrepalanchowk Makawanpur, Lalitpur	Fodder, Firewood, Insecticide (Malla & Chhetri, 2009; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Nagarkoti & Shrestha, 2022)
104	*Cinnamomum camphora* (L.) J.Presl Lauraceae	Nuwakot	Pneumonia (Nepali et al. 2020)
105	*Cinnamomum glanduliferum* (Wall.) Meisn. Lauraceae	Dolakha	Muscular swelling, Oil extraction (Karki et al., 2023)
106	*Cinnamomum glaucescens* (Nees) Hand.-Mazz. Lauraceae	Ramechaap, Makawanpur	Cough, Blood pressure, Sinusitis, Fever, Spice, Flavoring, Fodder, Diarrhea, Vomiting, Stomach problems, Oil extraction (Bhattarai & Tamang, 2017; Pradhan et al., 2020)
107	*Cinnamomum tamala* (Buch.-Ham.) T.Nees & C.H.Eberm. Lauraceae	Bhaktapur, Makawanpur, Ramechaap, Dolakha, Lalitpur, Sinduli, Kathmandu	Dyspepsia, Dysentery (Balami, 2004; Shah & Singh, 2014; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023; Moktan, 2023)
108	*Cinnamomum verum* J.Presl Lauraceae	Sinduli	Cough, Cold, Indigestion, Stomach problem, Bleeding gums, Fodder, Edible fruits, Toothbrush (twigs) (Shah & Singh, 2014)
109	*Cipadessa baccifera* Miq. Meliaceae	Ramechaap, Kavrepalanchowk	Fever, Typhoid, Throat pain, Urinary trouble, Placenta removal, Skin diseases, Jaundice, Neck pain, Infertility (Malla & Chhetri, 2009; Pradhan et al., 2020)
110	*Cirsium verutum* Spreng. Asteraceae	Lalitpur, Dolakha, Kathmandu	Energy tonic, Fever, Urine sensation (Balami, 2004; Panta, 2019; Dhital et al., 2021; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
111	*Cirsium wallichii* DC. Asteraceae	Makawanpur, Bhaktapur	Menstruation problem, Gynecological disorders, Post-delivery recovery, Milk production in animals, Abdominal cramping, Typhoid, Appetite loss, Blood clotting, Cough, Snake bites, Gastric issues, Ulcer, Burning sensation, Worms, Fever, Abortion (Joshi et al., 2020; Dulal et al., 2022)
112	*Cissampelos pareira* L. Menispermaceae	Chitwan, Lalitpur, Bhaktapur, Dolakha, Makawanpur, Kathmandu	Cataract, Corneal opacities, Edible fruits (Balami, 2004; Dangol & Gurung, 2008; Luitel et al., 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Bhaila et al., 2022; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
113	*Cissus adnata* Roxb. Vitaceae	Dolakha, Makawanpur	Boils, Bone fracture, Pickled leaves (Karki et al., 2023; Moktan, 2023)
114	*Cissus discolor* Blume Vitaceae	Makawanpur	Anemia, Dyspepsia, Fracture, Cholesterol control, Eye trouble, Rheumatism, Dysentery, Diarrhea, Indigestion, Blood pressure, Wounds (Joshi et al., 2020; Moktan, 2023)
115	*Citrus aurantiifolia* (Christm.) Swingle Rutaceae	Kavrepalanchowk, Makawanpur, Sidhuli, Ramechaap	General weakness, Impotence, Pimples, Skin ointment (Malla & Chhetri, 2009; Shah & Singh, 2014; Pradhan et al., 2020; Moktan, 2023)
116	*Citrus aurantium* L. Rutaceae	Kavrepalanchowk, Sinduli Bhaktapur	Antiseptic, Choked voice, Burning urination, Common cold, Indigestion, Vomiting, Anorexia (Malla & Chhetri, 2009; Shah & Singh, 2014; Dulal et al., 2022)
117	*Citrus limon* (L.) Osbeck Rutaceae	Sinduli, Dhading, Bhaktapur, Makawanpur, Sindupalchowk	Cough, Asthma, Weakness, Skin infection (Shah & Singh, 2014; Dulal et al., 2022; Gurung & Subedi, 2021; Gautam & Dhakal, 2023; Gurung, 2023)
118	*Citrus maxima* (Burm.) Merr. Rutaceae	Sinduli	Anthelmintic, Fever, Gastritis, Digestion aid, Dysentery (Shah & Singh, 2014)
119	*Citrus medica* L. Rutaceae	Chitwan, Lalitpur, Dolakha, Bhaktapur	Cough, Cold, Sinusitis, Headache (Poudel & Singh, 2016; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
120	*Clematis buchananiana* DC. Ranunculaceae	Ramechaap, Makawanpur, Dolakha, Sindupalchowk, Kathmandu	Cuts, Wounds (especially on children's heads) (Balami, 2004; Rai et al., 2004; Joshi et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
121	*Cleome viscosa* L. Capparadaceae	Chitwan	Agiya, Antihelmintic, Anti-lice, Toothache, Ulcer (Dangol & Gurung, 2008)
122	*Clerodendrum indicum* Kuntze Lamiaceae	Makawanpur	Blood pressure regulation, Blood purification, Menstrual cramps, Worms, Fever (Joshi et al., 2020)
123	*Clerodendrum infortunatum* L. Lamiaceae	Chitwan, Sinduli	Cough, Cold (Dangol & Gurung, 2008; Kafle & Dani, 2022)
124	*Clinopodium piperitum* (D.Don) Murata Lamiaceae	Makawanpur	Cuts, Wounds (Joshi et al., 2020)
125	*Clinopodium umbrosum* (M.Bieb.) K.Koch Lamiaceae	Makawanpur, Dolakha	Hair tonic (Joshi et al., 2020; Karki et al., 2023)
126	*Cocos nucifera* L. Arecaceae	Sinduli	Cuts (pseudobulb paste) (Kafle & Dani, 2022)
127	*Coelogyne corymbosa* Lindl. Orchidaceae	Dolakha	Boils, Cuts, Wounds (pseudobulb paste) (Karki et al., 2023)
128	*Coelogyne cristata* Lindl. Orchidaceae	Makawanpur, Dolakha	Stomach ache, Fever, Ornamental (necklace), Urinary troubles (Joshi et al., 2020; Karki et al., 2023)
129	*Coix lacryma-jobi* L. Poaceae	Makawanpur, Sinduli, Bhaktapur	Epilepsy, Harital (body weakness/eye problem), Fever, Headaches, Wounds, Gastric problems, Sinusitis, Fodder, Snake bite, Ear ache, Appetite (Bhaila et al., 2022; Kafle & Dani, 2022; Moktan, 2023)
130	*Colebrookea oppositifolia* Sm. Lamiaceae	Kavrepalanchowk, Makawanpur, Sindupalchowk, Chitwan, Dolakha	Dysentery, Lactation, Vegetable (leaves), Swollen body parts, Masaura preparation (dried shoots) (Rai et al., 2004; Dangol & Gurung, 2008; Malla & Chhetri, 2009; Bhattarai & Tamang, 2017; Joshi et al., 2020; Karki et al., 2023)
131	*Colocasia esculenta* (L.) Schott Araceae	Kavrepalanchowk, Makawanpur, Bhaktapur, Chitwan, Sinduli	Fever, Measles (Malla & Chhetri, 2009; Shah & Singh, 2014; Poudel & Singh, 2016; Dulal et al., 2022; Moktan, 2023)
132	*Corchorus aestuans* L. Tiliaceae	Makawanpur	Digestion (Moktan, 2023)
133	*Coriandrum sativum* L. Apiaceae	Bhaktapur, Makawanpur	Blisters (Bhattarai & Tamang, 2017; Joshi et al., 2020; Dulal et al., 2022)
134	*Crassocephalum crepidioides* S.Moore Asteraceae	Lalitpur	Urinary disorders, Appetite loss, Hypertension, Kidney stones, Cancer, Blood pressure
135	*Crateva unilocularis* Buch.-Ham. Capparaceae	Chitwan Nuwakot, Bhaktapur, Dhading	Mumps (Timilsina & Singh, 2015; Poudel & Singh, 2016; Gurung & Subedi, 2021; Bhaila et al., 2022)
136	*Crinum asiaticum* L.	Sinduli	Dhokre (Kafle & Dani, 2022)

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	Amaryllidaceae		
137	*Crotalaria prostrata* Roxb. Fabaceae	Sinduli	Dan, Hematuria (Kafle & Dani, 2022)
138	*Croton persimilis* Müll.Arg. Euphorbiaceae	Sinduli	Dysentery (Kafle & Dani, 2022)
139	*Cucumis melo* L. Cucurbitaceae	Chitwan	Heat stone problem, Cooling tonic (Dangol & Gurung, 2008)
140	*Cucumis sativus* L. Cucurbitaceae	Chitwan Bhaktapur	Jaundice, Pickle (seeds) (Poudel & Singh, 2016; Dulal et al., 2022)
141	*Cucurbita maxima* Duchesne Cucurbitaceae	Chitwan, Kavrepalanchowk	Jaundice (Malla & Chhetri, 2009; Poudel & Singh, 2016)
142	*Curculigo orchioides* Gaertn. Hypoxydaceae	Makawanpur	Bone fracture, Cough, Cold, Wounds, Throat (Bhattarai & Tamang, 2017; Joshi et al., 2020)
143	*Curcuma angustifolia* Roxb. Zingiberaceae	Ramechaap, Makawanpur	Food poisoning, Appetite stimulation, Sarki khatera, Evil eye, Piles, Placenta retention, Burning urination, Gastritis, Acidity, Blood purification, Skin diseases, Antidote (Bhattarai & Tamang, 2017; Pradhan et al., 2020)
144	*Curcuma caesia* Roxb. Zingiberaceae	Lalitpur, Dolakha, Sinduli, Bhaktapur, Sindupalchowk	Insect bites, Wounds, Allergies, Cough, Cold, Tonsillitis, Inflammation, Body pain, Mouth infection, Flavoring, Coloring, Fracture (in poultry) (Panta, 2019; Dulal et al., 2022; Kafle & Dani, 2022; Gurung, 2023; Karki et al., 2023)
145	*Curcuma longa* L. Zingiberaceae	Ramechaap, Kavrepalanchowk, Makawanpur, Sinduli, Dolakha, Laltipur, Bhaktapur, Sindupalchowk	Appetite, Body ache (Malla & Chhetri, 2009; Shah & Singh, 2014; Panta, 2019; Pradhan et al., 2020; Dhital et al., 2021; Dulal et al., 2022; Gurung, 2023; Moktan, 2023)
146	*Cuscuta cassytoides* Nees Convolvulaceae	Dolakha	Jaundice, High blood pressure, Blood purification, Asthma, Diarrhea, Fever, Urine blockage (Dhital et al., 2021)
147	*Cuscuta reflexa* Roxb. Convolvulaceae	Chitwan, Dolakha, Nuwakot, Lalitpur, Sinduli, Sundhupalchowk, Bhaktapur, Makawanpur, Kathmandu	Marcha preparation (fermenting agent) (Balami, 2004; Rai et al., 2004; Dangol & Gurung, 2008; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
148	*Cyanthillium cinereum* (L.) H.Rob. Asteraceae	Makawanpur	Nose bleeding, Body coolant, Jaundice, Urine problems, Albinism, Burning urination, Cuts, Wounds, Epilepsy, Fever, Hemorrhage, Stomachache, Hair removal, Herpes zoster, Melasma, Religious ceremonies, Cultural events(Moktan, 2023)
149	*Cynodon dactylon* (L.) Pers. Poaceae	Nuwakot, Chitwan, Bhaktapur, Lalitpur, Ramechaap, Sinduli Makawanpur, Dolakha, Dhading, Kathmandu	Cuts, Wounds, Boils, Diarrhea, Mumps (Balami, 2004; Rai et al., 2004; Dangol & Gurung, 2008; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Bhaila et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Karki et al., 2023; Moktan, 2023)
150	*Cynoglossum zeylanicum* (Sw. ex Lehm.) Thunb. ex Brand Boraginaceae	Lalitpur, Makawanpur, Nuwakot	Cuts, Scabies (Balami, 2004; Timilsina & Singh, 2015; Panta, 2019; Joshi et al., 2020; Nagarkoti & Shrestha, 2022)
151	*Cyperus compressus* L. Cyperaceae	Chitwan	Anthelmintic for livestock, Headache (Dangol & Gurung, 2008)
152	*Cyperus rotundus* L. Cyperaceae	Sinduli, Bhaktapur, Kathmandu	Snake bite, Burns, Cuts, Wounds, Diabetes, Dysentery, Diarrhea, Seminal weakness, Incense, Vegetable (young leaves) (Balami, 2004; Dulal et al., 2022; Kafle & Dani, 2022)
153	*Dactylorhiza hatagirea* (D.Don) Soó Orchidaceae	Ramechaap, Rasuwa, Nuwakot	Fever, Diarrhea, Dysentery, Cooling effect, Construction (heartwood), Fuelwood (Shrestha et al., 2014; Timilsina & Singh, 2015; Pradhan et al., 2020)
154	*Dalbergia sissoo* Roxb. ex DC. Fabaceae	Chitwan, Nuwakot, Sinduli, Ramechaap	Fever, Intestinal disorder, Parasites, Paper making, Animal stimulant, Rope making (Dangol & Gurung, 2008; Nepali et al., 2020; Pradhan et al., 2020; Kafle & Dani, 2022)
155	*Daphne bholua* Buch.-Ham. ex D.Don Thymelaeaceae	Kavrepalanchowk, Makawanpur	Dhokre, Earache, Itching (Malla & Chhetri, 2009; Bhattarai & Tamang, 2017)
156	*Datura metel* L. Solanaceae	Sinduli, Bhaktapur, Makawpur	Asthma, Cough, Toothaches, Fish bite, Body swelling, Cough in cattle, Rabies, Menstrual disorder (Bhattarai & Tamang, 2017; Dulal et al., 2022; Kafle & Dani, 2022)
157	*Datura stramonium* L. Solanaceae	Lalitpur, Dolakha, Sindupalchowk	Food poisoning (Rai et al., 2004; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
158	*Delphinium cooperi* Munz Ranunculaceae	Dhading, Sindupalchowk, Kathmandu	Cough, Diarrhea, Liver problems, Blood ailments (Balami, 2004; Gurung & Subedi, 2021; Gurung, 2023)
159	*Delphinium himalayae* Munz Ranunculaceae	Rasuwa	Skin disease, Ornamental (Shrestha et al, 2014)
160	*Dendrobium densiflorum* Lindl. Orchidaceae	Ramechaap, Makawanpur	Tonic (Joshi et al., 2020; Pradhan et al., 2020)
161	*Dendrobium longicornu* Lindl. Orchidaceae	Makawanpur	Body strength, Sexual desire, Vegetable (tender shoots), Fodder, Construction, Basket making, Flute making, Mat making (Joshi et al., 2020)
162	*Dendrocalamus hamiltonii* Nees & Arn. ex Munro Poaceae	Kavrepalanchowk, Chitwan, Makawanpur, Ramechaap	Stomach pain (Dangol & Gurung, 2008; Malla & Chhetri, 2009; Pradhan et al., 2020; Moktan, 2023)
163	*Desmostachya bipinnata* (L.) Stapf Poaceae	Sinduli	Fever (Kafle & Dani, 2022)
164	*Dichrocephala integrifolia* (L.f.) Kuntze Asteraceae	Bhaktapur (Suryabinak)	Cut wounds, Kidney stones (Bhaila et al., 2022)
165	*Didymocarpus pedicellatus* R.Br. Gesneriaceae	Makawanpur Sindupalchowk	Haarital treatment (with Curcuma caesia and Colebrookia oppositifolia) (Rai et al., 2004; Joshi et al., 2020)
166	*Dimetia scandens* (Roxb.) R.J.Wang Rubiaceae	Dolakha	Throat pain (Karki et al., 2023)
167	*Dioscorea alata* L. Dioscoreaceae	Dhading Bhaktapur	Chest pain, Piles, Dysentery, Syphilis, Ulcers, Appetite loss, Edible (boiled tubers) (Sharma & Bastakoti, 2009; Dulal et al., 2022)
168	*Dioscorea bulbifera* L. Dioscoreaceae	Dhading, Makawanpur, Kavrepalanchowk, Kathmandu	Abdominal pain, Antifertility (Balami, 2004; Malla & Chhetri, 2009; Joshi et al., 2020; Sharma & Bastakoti, 2010; Gurung & Subedi, 2021; Moktan, 2023)
169	*Dioscorea deltoidea* Wall. ex Griseb. Dioscoreaceae	Bhaktapur, Makawanpur, Dhading, Kathmandu	Edible (tubers and bulbils) (Balami, 2004; Sharma & Bastakoti, 2009; Bhattarai & Tamang, 2017; Joshi et al., 2020; Dulal et al., 2022; Moktan, 2023)
170	*Dioscorea esculenta* (Lour.) Burkill Dioscoreaceae	Dhading	Sooth voice, Cough, Fever (Sharma & Bastakoti, 2009)
171	*Dioscorea hamiltonii* Hook.f.	Dhading, Dolakha	Harinad (peeling skin on feet), Diuretic, Edible (bulbils and tubers), Poison antidote (Sharma & Bastakoti, 2009; Karki et al., 2023)

S. N.	Species/Family	Reported districts	Uses
	Dioscoreaceae		
172	*Dioscorea hispida* Dennst. Dioscoreaceae	Dhading	Edible (bulbils and tubers), Poisoning antidote (Sharma & Bastakoti, 2009)
173	*Dioscorea kamoonensis* Kunth Dioscoreaceae	Dhading	Boils, Vegetable (tubers) (Sharma & Bastakoti, 2009)
174	*Dioscorea pentaphylla* L. Dioscoreaceae	Kavrepalanchowk, Dhading	Edible (bulbils and tubers) (Malla & Chhetri, 2009; Sharma & Bastakoti, 2009)
175	*Dioscorea pubera* Blume Dioscoreaceae	Dhading	Intestinal worms (Sharma & Bastakoti, 2009)
176	*Diplazium esculentum* (Retz.) Sw. Athyriaceae	Bhaktapur, Makawanpur	Cuts, Wounds, Livestock bedding, Manure preparation (Bhaila et al., 2022; Moktan, 2023)
177	*Diplazium polypodioides* Blume Athyriaceae	Kavrepalanchowk	Diarrhea, Dysentery, Vegetable (tender shoots), Manure preparation (Malla & Chettri , 2009)
178	*Diplazium stoliczkae* Bedd. Athyriaceae	Kavrepalanchowk	Edible fruits, Vegetable ghee (seeds), Rheumatism, Foot cracks, Wounds (Malla & Chettri , 2009)
179	*Diploknema butyracea* (Roxb.) H.J.Lam Sapotaceae	Makawanpur, Nuwakot, Dhading	Ulcers, Snake repellent (Joshi et al., 2020; Nepali et al., 2020; Gurung & Subedi, 2021; S. Bhattarai et al., 2024)
180	*Dracaena trifasciata* (Prain) Mabb. Agavaceae	Chitwan	Cough, Cold, Indigestion, Sinusitis, Burns, Fever, Snake bites (Dangol & Gurung, 2008)
181	*Dracocephalum heterophyllum* Benth. Lamiaceae	Ramechaap	Piles, Vegetable (young shoots), Pickle, Tama (fermented), Basket making, Mat making, Fishing rods, Ceiling, Fodder (Pradhan et al., 2020)
182	*Drepanostachyum falcatum* (Nees) Keng f. Poaceae	Lalitpur, Kavrepalanchowk	Fodder, Weaving mats, Baskets, Walking sticks, Vegetable (young shoots), Pickle (Malla & Chhetri, 2009; Panta, 2019)
183	*Drepanostachyum intermedium* (Munro) Keng f. Poaceae	Ramechaap, Kavrepalanchowk	Headache, High blood pressure (Malla & Chhetri, 2009; Pradhan et al., 2020)
184	*Drimia indica* (Roxb.) Jessop Asparagaceae	Sinduli	Fever, Common cold, Conjunctivitis, Sinusitis, Headache, Gastritis, Indigestion, Appetite loss, Skin inflammation, Urine inflammation, Ringworm, Scabies, Cut wounds, Typhoid, Fodder (Kafle & Dani, 2022)
185	*Drymaria cordata* Willd. ex Schult. Caryophyllaceae	Chitwan, Dolakha, Makawanpur, Bhaktapur, Lalitpur, Kathmandu	Common cold, Sinusitis, White pupil (Balami, 2004; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Dulal et al., 2022; Karki et al., 2023; Moktan, 2023)
186	*Drymaria villosa* Cham. & Schltdl. Caryophyllaceae	Lalitpur	Cattle bedding (Nagarkoti & Shrestha, 2022)
187	*Dryopteris sparsa* (D.Don) Kuntze Dryopteridaceae	Makawanpur	Anthelmintic, Insect repellent (Moktan, 2023)
188	*Duranta erecta* L. Verbenaceae	Bhaktapur	Cuts, Wounds, Headache, Athlete's foot (Dulal et al., 2022)
189	*Eclipta prostrata* (L.) L. Asteraceae	Chitwan, Sinduli, Makawanpur, Kathmandu	Fracture (Balami, 2004; Dangol & Gurung, 2008; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Kafle & Dani, 2022)
190	*Elaeagnus infundibularis* Momiy. Elaeagnaceae	Dolakha	Liver tonic, Blood pressure, Mental disorders, Religious significance, Ornamental (seeds) (Karki et al., 2023)
191	*Elaeocarpus angustifolius* Blume Elaeocarpaceae	Kavrepalanchowk	Wounds treatment (especially waterlogged) (Malla & Chettri , 2009)
192	*Elephantopus scaber* L. Asteraceae	Makawanpur, Sinduli	Cough, Cold, Fever, Hair growth, Marcha preparation (fermenting cake) (Luitel et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Kafle & Dani, 2022; Moktan, 2023)
193	*Eleusine coracana* (L.) Gaertn. Poaceae	Sinduli	Body pain, Relaxation (Kafle & Dani, 2022)
194	*Elsholtzia blanda* Benth. Lamiaceae	Makawanpur, Dhading	Fever, Pneumonia, Chest pain, Sinusitis (Joshi et al., 2020; Gurung & Subedi, 2021; Moktan, 2023)
195	*Elsholtzia flava* Benth. Lamiaceae	Dolakha, Kathmandu	Bone fracture (Balami, 2004; Karki et al., 2023)
196	*Elsholtzia fruticosa* Rehder Lamiaceae	Sinduli	Skin infection (Kafle & Dani, 2022)
197	*Engelhardia spicata* Lechen ex Blume Juglandaceae	Sindupalchowk, Sinduli	Skin diseases, Burns (Rai et al., 2004; Shah & Singh, 2014)
198	*Ephedra gerardiana* Wall. ex Klotzsch & Garcke Ephedraceae	Rasuwa, Makawanpur	Asthma, Respiratory diseases, Cardiac stimulant (Shrestha et al., 2014; Bhattarai & Tamang, 2017)
199	*Equisetum arvense* L. Equisetaceae	Dolakha	Urine blockage (Karki et al., 2023)
200	*Equisetum diffusum* D.Don Equisetaceae	Sindupalchowk Ramechaap	Jaundice, Fever, Eye problems, Cough, Cold (Rai et al., 2004; Pradhan et al., 2020)
201	*Equisetum ramosissimum* Desf. Equisetaceae	Makawanpur	Scabies, Burns, Urinary tract infection (Joshi et al., 2020)
202	*Eragrostis tenella* (L.) P.Beauv. ex Roem. & Schult. Poaceae	Makawanpur	Fodder (Moktan, 2023)
203	*Eriocapitella vitifolia* (Buch.-Ham. ex DC.) Nakai Ranunculaceae	Ramechaap, Makawanpur	Scabies, Stomach problems, Headache, Mental disorder, Diarrhea, Vomiting, Constipation, Vegetable (shoots), Spice (Joshi et al., 2020; Pradhan et al., 2020)
204	*Erythrina stricta* Roxb. Fabaceae	Dhading, Makawanpur	Diarrhea, Body coolant (Bhattarai & Tamang, 2017; Gurung & Subedi, 2021)
205	*Eulaliopsis binata* (Retz.) C.E.Hubb. Poaceae	Kavrepalanchowk, Makawanpur	Rope making, Homemade paper, Thatching roofs, Fodder (Malla & Chettri, 2009; Bhattarai & Tamang, 2017; Moktan, 2023)
206	*Eupatorium chinense* L. Asteraceae	Rasuwa, Kathmandu	Skin infection (Balami, 2004; Shrestha et al., 2014)
207	*Euphorbia hirta* L. Euphorbiaceae	Chitwan, Kavrepalanchowk, Makawanpur, Sinduli, Bhaktapur, Kathmandu	Numb legs, Cough, Diarrhea, Dysentery, Asthma, Bronchial infections, Wounds, Dislocated bones, Snake bites, Milk production, Boils, Earache, Hypertension, Diabetes, Lactation (Balami, 2004; Dangol & Gurung, 2008; Malla & Chhetri, 2009; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Bhaila et al., 2022; Kafle & Dani, 2022)

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208	*Euphorbia pulcherrima* Willd. ex Klotzsch Euphorbiaceae	Lalitpur, Kavrepalanchowk	Boils, Skin diseases, Toothache, Ornamental, Ritual purposes (Malla & Chhetri, 2009; Panta, 2019)
209	*Euphorbia royleana* Boiss. Euphorbiaceae	Makawanpur, Sinduli, Chitwan, Bhaktapur	Common cold, Cough, Dysentery, Fever, Bleeding, Appetite, Sinusitis, Toothache (Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Dulal et al., 2022; Kafle & Dani, 2022; Moktan, 2023)
210	*Euphorbia thymifolia* L. Euphorbiaceae	Sindupalchowk	Cut wounds (Rai et al., 2004)
211	*Exallage ulmifolia* (Wall.) Bremek. Rubiaceae	Makawanpur	Stomachache (Joshi et al., 2020)
212	*Fagopyrum acutatum* Mansf. ex K.Hammer Polygonaceae	Dhading	Body coolant (Gurung & Subedi , 2021)
213	*Fagopyrum esculentum* Moench Polygonaceae	Chitwan	Typhoid (Poudel & Singh, 2016)
214	*Ficus auriculata* Lour. Moraceae	Makawanpur, Bhaktapur	Diarrhea, Fodder, Edible (ripe figs) (Bhaila et al., 2022; Moktan, 2023)
215	*Ficus benghalensis* L. Moraceae	Kavrepalanchowk, Chitwan, Dolakha, Ramechaap, Makawanpur	Diarrhea, Dysentery, Hair problems, Gastritis, Pains, Bruises, Rheumatism, Toothache, Abscesses, Corneal opacities, Religious ceremonies, Fodder, Timber, Fuelwood, Edible fruits (Malla & Chhetri, 2009; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Karki et al., 2023)
216	*Ficus benjamina* L. Moraceae	Ramechaap	Boils, Cuts, Wounds, Religious ceremonies, Timber (Pradhan et al., 2020)
217	*Ficus glaberrima* Blume Moraceae	Kavrepalanchowk	Edible fruits, Rope making, Fodder (Malla & Chettri , 2009)
218	*Ficus hispida* L.f. Moraceae	Makawanpur, Chitwan, Nuwakot	Fodder, Firewood, Earache, Hearing weakness, Edible fruits (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Nepali et al., 2020; Moktan, 2023)
219	*Ficus lacor* Buch.-Ham. Moraceae	Makawanpur, Nuwakot	Boils, Fodder, Vegetable (buds), Pickle (Joshi et al., 2020; Nepali et al., 2020)
220	*Ficus racemosa* L. Moraceae	Dhading, Makawanpur, Nuwakot	Cuts, Wounds, Fodder, Edible fruits, Fuelwood (Bhattarai & Tamang, 2017; Nepali et al., 2020; Gurung & Subedi, 2021; Moktan, 2023)
221	*Ficus religiosa* L. Moraceae	Chitwan, Sinduli, Lalitpur, Ramechaap, Bhaktapur	Strain, Muscle crack, Breaks, Skin disease, Headache, Boils, Piles, Stomachache, Constipation, Menstrual regulation, Religious ceremonies, Fodder, Timber, Fuelwood (Dangol & Gurung, 2008; Shah & Singh, 2014; Panta, 2019; Pradhan et al., 2020; Dulal et al., 2022; Nagarkoti & Shrestha, 2022)
222	*Ficus semicordata* Buch.-Ham. ex Sm. Moraceae	Ramechaap, Makawanpur, Nuwakot, Dhading, Bhaktapur	Leprosy, Fever, Bone fracture, Cuts, Laxative, Fodder, Edible fruits, Rope making, Fuelwood (Bhattarai & Tamang, 2017; Nepali et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Moktan, 2023)
223	*Flemingia macrophylla* (Willd.) Kuntze ex Merr. Fabaceae	Makawanpur	Fodder (Moktan, 2023)
224	*Flemingia strobilifera* (L.) W.T.Aiton Fabaceae	Sinduli	Dysentery, Diarrhea (Kafle & Dani, 2022)
225	*Foeniculum vulgare* Mill. Umbelliferae	Nuwakot	Chest, Kidney, Spleen trouble (Timilsina & Singh, 2015)
226	*Fragaria nubicola* Lindl. ex Lacaita Rosaceae	Rasuwa	Profuse menstruation, Edible fruits (Shrestha et al, 2014)
227	*Fraxinus floribunda* Wall. Oleaceae	Dolakha	Fracture (Karki et al., 2023)
228	*Fritillaria cirrhosa* D.Don Liliaceae	Rasuwa	Stomach pain, Gastritis (Shrestha et al, 2014)
229	*Gardenia jasminoides* J.Ellis Rubiaceae	Chitwan	Thirst quencher (Dangol & Gurung, 2008)
230	*Garuga pinnata* Roxb. Anacardiaceae	Kavrepalanchowk	Pulmonary affections, Skin diseases, Dislocated bones, Wounds, Asthma, Roundworm, Fodder (Malla & Chettri , 2009)
231	*Gaultheria fragrantissima* Wall. Ericaceae	Lalitpur, Makawanpur, Dolakha, Nuwakot, Ramechaap, Kathmandu	Body pain, Joint pain, Cough, Cold, Anti-alcoholic, Rheumatism, Anthelmintic, Antiseptic, Stomach disorder, Diarrhea, Scabies, Edible fruits, Essential oil extraction (Balami, 2004; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Gautam & Dhakal, 2023; Karki et al., 2023)
232	*Gaultheria nummularioides* D.Don Ericaceae	Makawanpur	Urine sensation (Joshi et al., 2020)
233	*Girardinia diversifolia* (Link) Friis Urticacae	Makawanpur, Ramechaap, Dhaading, Kathmandu	Constipation, Headaches, Joint pain, Tonic, Vegetable (young leaves), Cloth making (Balami, 2004; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021)
234	*Globba racemosa* Sm. Zingiberaceae	Makawanpur Ramechaap	Stomach problems, Urinary infections, Headache, Body pain Joshi et al., 2020; Pradhan et al., 2020)
235	*Glycyrrhiza glabra* L. Fabaceae	Sinduli, Makawanpur	Tonsillitis (Bhattarai & Tamang, 2017; Kafle & Dani, 2022)
236	*Gmelina arborea* Roxb. Verbenaceae	Chitwan	Swollen necks of bullocks (Dangol & Gurung, 2008)
237	*Gonostegia triandra* (Blume) Miq. Urticacae	Bhaktapur, Lalitpur	Dislocated bone treatment (Bhaila et al., 2022; Dulal et al., 2022)
238	*Gossypium* L. Malvaceae	Sindupalchowk	Fever (Rai et al., 2004)
239	*Grewia sapida* Roxb. ex DC. Tiliaceae	Chitwan	Dysentery (Dangol & Gurung, 2008)
240	*Hedychium ellipticum* Buch.-Ham. ex Sm. Zingiberaceae	Nuwakot	Fever (Timilsina & Singh, 2015)
241	*Helianthus annuus* L. Asteraceae	Sinduli, Nuwakot	Blood pressure, Lung infection, Ulcers, Wounds, Burnt skin (Shah & Singh, 2014; Timilsina & Singh, 2015)
242	*Helicteres isora* L. Sterculiaceae	Chitwan	Cattle fever, Roundworm infection (Dangol & Gurung, 2008)
243	*Hellenia speciosa* (J.Koenig) S.R.Dutta Costaceae	Sinduli, Makawanpur, Chitwan	Burning urination, Fever, Joint pain, Thirst (Poudel & Singh, 2016; Kafle & Dani, 2022; Moktan, 2023)
244	*Hemionitis albomarginata* (C.B.Clarke)	Bhaktapur, Makawanpur	Gastritis, Ulcer, Stomach problems (Bhaila et al., 2022; Moktan, 2023)

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	Christenh. Pteridaceae		
245	*Hemionitis bicolor* (Roxb.) Christenh. Pteridaceae	Sinduli	Fever, Cooling agent (Kafle & Dani, 2022)
246	*Hemiphragma heterophyllum* Wall. Plantaginaceae	Dolakha	Stomach disorder, Constipation, Gano-gola (Karki et al., 2023)
247	*Heynea trijuga* Roxb. Meliaceae	Chitwan, Makawanpur, Dolakha	Anti-lice, Dermatological allergies, Menstruation disorders (Dangol & Gurung, 2008; Joshi et al., 2020; Karki et al., 2023)
248	*Hibiscus rosa-sinensis* L. Malvaceae	Sinduli, Ramechaap	Body cooling, Skin infection, Blood pressure, Urinary problems, Excess bleeding, Ornamental, Religious ceremonies (Shah & Singh, 2014; Pradhan et al., 2020)
249	*Hibiscus sabdariffa* L. Malvaceae	Sinduli	Blood purification in women (Kafle & Dani, 2022)
250	*Himalaiella deltoidea* (DC.) Raab-Straube Asteraceae	Makawanpur	Fever (Joshi et al., 2020)
251	*Hippophae salicifolia* D.Don Elaeagnaceae	Rasuwa	Tonic, Diarrhea (Shrestha et al, 2014)
252	*Hippophae tibetana* Schltdl. Elaeagnaceae	Rasuwa	Tonic, Diarrhea (Shrestha et al, 2014)
253	*Holarrhena pubescens* Wall. & G.Don Apocynaceae	Chitwan, Makawanpur	Heat (Poudel & Singh, 2016; Bhattarai & Tamang, 2017)
254	*Houttuynia cordata* Thunb. Saururacae	Dolakha	Fever, Gastrointestinal disorders (Karki et al., 2023)
255	*Hydnum repandum* L. Hydnaceae	Makawanpur	Vegetable, Pickle (Moktan, 2023)
256	*Hydrangea febrifuga* (Lour.) Y.De Smet & Granados Hydrangiaceae	Dolakha	High fever (Karki et al., 2023)
257	*Hydrocotyle javanica* Thunb. Araliaceae	Dolakha, Kathmandu	Snake bite (Balami, 2004; Dhital et al., 2021)
258	*Hydrocotyle modesta* Cham. & Schltdl. Araliaceae	Dolakha	Urine blockage (Karki et al., 2023)
259	*Hydrocotyle rotundifolia* Roxb. Araliaceae	Chitwan	Fever, Urinary disorders (Dangol & Gurung, 2008)
260	*Hydrocotyle sibthorpioides* Lam. Araliaceae	Lalitpur	Irritation in children (Nagarkoti & Shrestha, 2022)
261	*Hypericum cordifolium* Choisy Hypericaceae	Chitwan	Rheumatism (Poudel & Singh, 2016)
262	*Hypericum elodeoides* Choisy Hypericaceae	Dolakha	Cuts, Wounds (Dhital et al., 2021)
263	*Hypericum perforatum* L. Hypericaceae	Dhading	Burns (Gurung & Subedi , 2021)
264	*Hypericum uralum* Buch.-Ham. ex D.Don Hypericaceae	Nuwakot, Sindupalchowk	Cuts, Old wounds (Rai et al., 2004; Timilsina & Singh, 2015)
265	*Hyptianthera stricta* (Roxb. ex Sm.) Wight & Arn. Rubiaceae	Makawanpur	Typhoid (Joshi et al., 2020)
266	*Hyptis suaveolens* (L.) Poit. Lamiaceae	Makawanpur	Wounds (Joshi et al., 2020)
267	*Ichnocarpus frutescens* (L.) W.T.Aiton Apocynaceae	Sinduli	Agalactia, Milk production (Kafle & Dani, 2022)
268	*Impatiens balsamina* L. Balsaminaceae	Sinduli	Urinary problem (Shah & Singh, 2014)
269	*Imperata cylindrica* (L.) Raeusch. Poaceae	Dolakha, Ramechaap, Bhaktapur, Makawanpur, Kathmandu	Worms, Fever, Diarrhea, Fodder (Balami, 2004; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Dulal et al., 2022; Karki et al., 2023)
270	*Indigofera atropurpurea* Buch.-Ham. ex Hornem. Fabaceae	Makawanpur	Heating effect (Moktan, 2023)
271	*Inula cappa* (Buch.-Ham. ex D.Don) DC. Asteraceae	Makawanpur, Dolakha, Kathmandu	Indigestion, Stomachache, Dysentery, Fracture (Balami, 2004; Bhattarai & Tamang, 2017; Joshi et al., 2020; Karki et al., 2023)
272	*Ipomoea aquatica* Forssk. Convolvulaceae	Chitwan	Lactation (Poudel & Singh, 2016)
273	*Ipomoea batatas* (L.) Lam. Convolvulaceae	Sinduli	Sexual stimulant, Tonic (Shah & Singh, 2014)
274	*Ipomoea quamoclit* L. Convolvulaceae	Makawanpur	Detergent for bathing (Moktan, 2023)
275	*Iresine diffusa* Humb. & Bonpl. ex Willd. Amaranthaceae	Sinduli	Cut, Wound (Kafle & Dani, 2022)
276	*Ixeridium sagittarioides* (C.B Clarke) Pak & Kawana Asteraceae	Dolakha	Ear problems (Karki et al., 2023)
277	*Jasminum mesnyi* Hance Oleaceae	Bhaktapur	Tonsil, Blood pressure reduction, Red rashes (Dulal et al., 2022)
278	*Jatropha curcas* L. Euphorbiaceae	Nuwakot, Chitwan, Dhading, Ramechaap, Sinduli, Sindupalchowk, Makawanpur	Inflammation, Boils, Wound, Tooth problems, Poisoning, Cough, Fever, Asthma, Constipation, Athlete's foot, Foot/hand cracks and fissures, Oil extraction, Soil erosion control (Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Gurung & Subedi, 2021; Kafle & Dani, 2022; Gurung, 2023)
279	*Juglans regia* L. Juglandaceae	Chitwan, Dhading, Sindupalchowk, Dolakha, Bhaktapur, Makawanpur	Anthelmintic, Tooth problems, Skin allergies, Liver cirrhosis, Stomach parasites, Typhoid (Rai et al., 2004; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Gurung & Subedi, 2021; Dulal et al., 2022; Karki et al., 2023)
280	*Juniperus indica* Bertol. Cupressaceae	Kavrepalanchowk, Makawanpur	Appetizer, Diarrhea, Abdominal pain, Spleen diseases, Tumors, Piles, Bronchitis, Vaginal diseases, Toothache, Religious ceremonies (Malla & Chettri, 2009; Bhattarai & Tamang, 2017)

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281	*Juniperus recurva* Buch.-Ham. ex. D.Don Cupressaceae	Rasuwa, Makawanpur	Throat infection (Shrestha et al., 2014; Bhattarai & Tamang, 2017)
282	*Jurinea dolomiaea* Boiss. Asteraceae	Rasuwa	Diarrhea (Shrestha et al, 2014)
283	*Justicia adhatoda* L. Acanthaceae	Makawanpur, Nuwakot, Lalitpur, Sinduli, Bhaktapur, Chitwan	Malaria, Serious fever, Bronchitis, Dandruff, Common cold, Cough, Dislocated bones, Stomach pain, Joint leg pain, Bone fracture, Itching, Blood pressure (Balami, 2004; Dangol & Gurung, 2008; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Moktan, 2023)
284	*Kaempferia rotunda* L. Zingiberaceae	Dolakha	Fracture, Pneumonia (Karki et al., 2023)
285	*Kalanchoe pinnata* (Lam.) Pers. Crassulaceae	Dolakha	Jaundice, Kidney stone (Karki et al., 2023)
286	*Lablab purpureus* (L.) Sweet Fabaceae	Sinduli, Kavrepalanchowk, Bhaktapur	Fungal infection, Skin scars, Allergy, Stomachic (Malla & Chhetri, 2009; Dulal et al., 2022; Kafle & Dani, 2022)
287	*Lagenaria siceraria* (Molina) Standl. Cucurbitaceae	Kavrepalanchowk, Sinduli	Burns, Indigestion, Ulcers, Stomach acidity, Earache, Laxative, Jaundice, Constipation, Cooling, Vegetable (fruits), Emetic, Purgative (Malla & Chhetri, 2009; Shah & Singh, 2014; Kafle & Dani, 2022)
288	*Lagerstroemia parviflora* Roxb. Lythraceae	Makawanpur, Nuwakot	Fodder, Firewood, Construction (Nepali et al., 2020; Moktan, 2023)
289	*Lannea coromandelica* (Houtt.) Merr. Anacardiaceae	Kavrepalanchowk, Chitwan	Nosebleed, Ulcers, Fodder, Tannin, Fuel, Furniture, Construction (Dangol & Gurung, 2008; Malla & Chhetri, 2009)
290	*Lawsonia inermis* L. Lythraceae	Sinduli	Athletes foot, Hair color (Kafle & Dani, 2022)
291	*Lepidium sativum* L. Brassicaceae	Kavrepalanchowk, Makawanpur	Asthma, Cough, Bleeding piles, Syphilis, Pain relief, Vegetable (Malla & Chettri, 2009; Bhattarai & Tamang, 2017)
292	*Lepisorus mehrae* Fraser-Jenk. Polypodiaceae	Ramechaap	Back pain, Stomach problems, Fever, Pesticide, Animal diarrhea, Mouth wounds (Pradhan et al., 2020)
293	*Leptodermis lanceolata* Wall. Rubiaceae	Ramechaap	Broken bones, Cuts, Wounds, Additive for snails (Pradhan et al., 2020)
294	*Leptodesmia microphylla* (Thunb.) H.Ohashi & K.Ohashi Fabaceae	Makawanpur	Boils, Wounds (Joshi et al., 2020)
295	*Leucaena leucocephala* (Lam.) de Wit Fabaceae	Nuwakot	Firewood (Nepali et al. 2020)
296	*Leucas aspera* Link Lamiaceae	Chitwan	Ringworm (Dangol & Gurung, 2008)
297	*Leucas cephalotes* Spreng. Lamiaceae	Chitwan, Bhaktapur, Sinduli, Makawanpur	Ringworm, Body blisters, Sinusitis, Scabies, Stye/Blepharitis (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Bhaila et al., 2022; Kafle & Dani, 2022)
298	*Leucosceptrum canum* Sm. Lamiaceae	Dolakha	Fracture, Intestinal disorders, Throat pain (Dhital et al., 2021; Karki et al., 2023)
299	*Lichen* Lichen	Dolakha	Cuts, Wounds (Dhital et al., 2021)
300	*Ligusticopsis wallichiana* (DC.) Pimenov & Kljuykov Apiaceae	Makawanpur	Fever, Body pain (Joshi et al., 2020)
301	*Lindera neesiana* (Wall. ex Nees) Kurz Lauraceae	Ramechaap, Makawanpur, Sinduli, Dolakha, Nuwakot, Lalitpur	Gastric problems, Diarrhoea, Abdominal pain, Fever, Cough, Cold, Sexual weakness, Blood pressure, Gastritis, Abscess, Antitoxic, Veterinary use (Luitel et al., 2014; Shah & Singh, 2014; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
302	*Litchi chinensis* Sonn. Sapindaceae	Nuwakot	Edible fruits (Nepali et al. 2020)
303	*Litsea cubeba* (Lour.) Pers. Lauraceae	Lalitpur	Gastritis, Stomach pain, Spice, Flavoring
304	*Litsea glutinosa* (Lour.) C.B.Rob. Lauraceae	Makawanpur	Bone dislocation, Bone fracture (Joshi et al., 2020; Moktan, 2023)
305	*Litsea monopetala* (Roxb.) Pers. Lauraceae	Kavrepalanchowk, Makawanpur, Nuwakot, Sindupalchowk	Diarrhoea, Pain relief, Fodder, Fuel (Rai et al., 2004; Malla & Chhetri, 2009; Nepali et al., 2020; Moktan, 2023)
306	*Lobelia pyramidalis* Wall. Campanulaceae	Makawanpur, Dolakha, Sindupalchowk, Bhaktapur, Kathmandu	Fever, Fertility enhancement (Balami, 2004; Rai et al., 2004; Bhattarai & Tamang, 2017; Joshi et al., 2020; Dulal et al., 2022; Karki et al., 2023)
307	*Lonicera angustifolia* var. *myrtillus* (Hook.f. & Thomson) Q.E.Yang, Landrein, Borosova & Osborne Caprifoliaceae	Rasuwa	Stomach disorder (Shrestha et al, 2014)
308	*Lycopodium clavatum* L. Lycopodiaceae	Kavrepalanchowk, Ramechaap, Sindupalchowk, Makawanpur	Fever, Headache, Wound, Urinary problems, Skin disease, Fissures, Cracks, Rheumatism, Ceremonial use (Rai et al., 2004; Malla & Chhetri, 2009; Bhattarai & Tamang, 2017; Pradhan et al., 2020)
309	*Lycopodium japonicum* Thunb. Lycopodiaceae	Makawanpur, Dolakha	Cuts, Wounds, Boils, Crack feet, Jaundice (Bhattarai & Tamang, 2017; Joshi et al., 2020; Karki et al., 2023)
310	*Lygodium japonicum* (Thunb.) Sw. Lygodiaceae	Chitwan	Ringworm (Poudel & Singh, 2016)
311	*Lyonia ovalifolia* (Wall.) Drude Ericaceae	Dhading, Makawanpur, Dolakha, Nuwakot, Lalitpur, Rasuwa, Sindupalchowk, Bhaktapur, Kathmandu	Wounds, Scabies, Itching, Allergies, Skin diseases, Cloven hoof suppuration, Timber, Firewood (Balami, 2004; Rai et al., 2004; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Nepali et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
312	*Machilus duthiei* King ex Hook.f. Lauraceae	Makawanpur	Bone fracture, Skin disease (Joshi et al., 2020)
313	*Machilus odoratissimus* Nees Lauraceae	Ramechaap	Wounds, Allergy, Dislocated bones, Rodent control, Fodder (Pradhan et al., 2020)
314	*Macrotyloma uniflorum* (Lam.) Verdc. Fabaceae	Sinduli, Dhading, Bhaktapur, Ramechaap, Sindupalchowk,	Diabetes, Urinary tract stones, Chicken pox, Kidney stones, Vegetable (Bhattarai & Tamang, 2017; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gurung, 2023)

S. N.	Species/Family	Reported districts	Uses
		Makawanpur	
315	*Madhuca longifolia* (L.) J.F.Macbr. Sapotaceae	Kavrepalanchowk, Makawanpur	Boils, Diabetes, Cough, Cold, Rheumatism, Skin disease, Fish poison, Construction, Furniture, Fuel, Edible flowers, Alcohol production (Malla & Chhetri, 2009; Moktan, 2023)
316	*Maesa chisia* D.Don Primulaceae	Makawanpur, Bhhaktapur, Kathmandu	Insecticide, Ringworm (Balami, 2004; Joshi et al., 2020; Dulal et al., 2022)
317	*Magnolia champaca* (L.) Baill. ex Pierre Magnoliaceae	Nuwakot	Fodder, Religious purposes (Nepali et al. 2020)
318	*Maianthemum purpureum* (Wall.) LaFrankie Asparagaceae	Sinduli	Tonsillitis (Kafle & Dani, 2022)
319	*Mallotus philippensis* (Lam.) Müll.Arg. Euphorbiaceae	Chitwan, Makawanpur, Sinduli, Dhading	Abdominal disorder, Body pain, Gastritis, Toe wounds (Dangol & Gurung, 2008; Joshi et al., 2020; Gurung & Subedi, 2021; Kafle & Dani, 2022; Moktan, 2023)
320	*Mangifera indica* L. Anacardiaceae	Sinduli, Kavrepalanchowk, Chitwan, Makawanpur, Nuwakot	Dysentery, Vomiting, Jaundice, Rheumatism, Ulcer, Cough, Cold, Scabies, Skin disease, Edible fruits, Pickle, Religious, Furniture, Fuel (Malla & Chhetri, 2009; Luitel et al., 2014; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Nepali et al., 2020; Kafle & Dani, 2022; Moktan, 2023)
321	*Manihot esculenta* Crantz Euphorbiaceae	Makawanpur	Edible tuberous roots (Moktan, 2023)
322	*Melanochyla caesia* (Blume) Ding Hou Anacardiaceae	Dolakha	Diarrhea, Dysentery (Karki et al., 2023)
323	*Melastoma malabathricum* L. Melastomataceae	Makawanpur	Edible fruits (Moktan, 2023)
324	*Melia azedarach* L. Meliaceae	Lalitpur, Dhading, Kavrepalanchowk, Sinduli, Nuwakot, Sindupalchowk, Chitwan, Bhaktapur, Makawanpur, Kathmandu	Anthelminthic, Antidandruff, Leprosy, Scrofula, Skin diseases, Hysteria, Headache, Rheumatic pain, Fever, Lice control, Furniture, Fuel, Fodder (Balami, 2004; Rai et al., 2004; Dangol & Gurung, 2008; Malla & Chhetri, 2009; Bhattarai & Tamang, 2017; Panta, 2019; Nepali et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gurung, 2023)
325	*Melothria heterophylla* (Lour.) Cogn. Cucurbitaceae	Chitwan, Nuwakot	Hydrocele, Neck ache, Fever (Dangol & Gurung, 2008; Timilsina & Singh, 2015)
326	*Mentha* × *piperita* L. Lamiaceae	Lalitpur, Bhaktapur	Cold, Sore throat, Cough, Asthma, Sinusitis (Dulal et al., 2022; Nagarkoti & Shrestha, 2022)
327	*Mentha arvensis* L. Lamiaceae	Bhaktapur, Sindupalchowk, Makawanpur	Fever, Headache (Bhattarai & Tamang, 2017; Dulal et al., 2022; Gurung, 2023)
328	*Mentha spicata* L. Lamiaceae	Makawanpur, Dolakha, Lalitpur, Kavrepalanchowk, Bhaktapur	Tongue boils, Digestive issues, Fever, Jaundice, Nausea, Diarrhoea, Dysentery, Vomiting, Kidney stone, Gastric disorder, Pickle (Malla & Chhetri, 2009; Joshi et al., 2020; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Karki et al., 2023)
329	*Mikania micrantha* Kunth Asteraceae	Chitwan	Cuts, Wounds (Poudel & Singh, 2016)
330	*Millettia extensa* (Benth.) Benth. ex Baker Fabaceae	Makawanpur	Exoparasite removal, Skin diseases (Bhattarai & Tamang, 2017; Joshi et al., 2020)
331	*Millettia fruticosa* (DC.) Benth. ex Baker Fabaceae	Makawanpur	Skin diseases (Joshi et al., 2020)
332	*Millettia glaucescens* K*URZ* Fabaceae	Sinduli	Paralysis (Kafle & Dani, 2022)
333	*Mimosa pudica* L. Fabaceae	Chitwan, Makawanpur, Sinduli, Dhading, Ramechaap, Nuwakot,	Scorpion sting, Rheumatism, Diarrhea, Dysentery, Fever, Toothache, Cuts, Wounds, Diabetes, Boils, Gastritis, Joint pain, Stomach problems, Skin infection, Throat ache, Cooling (Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Kafle & Dani, 2022; Moktan, 2023)
334	*Mimosa rubicaulis* lam. Fabaceae	Ramechaap, Makawanpur	Skin problems, Wound healing, Strain, Fever, Urinary disorder, Fodder (Bhattarai & Tamang, 2017; Pradhan et al., 2020)
335	*Mirabilis jalapa* L. Nyctaginaceae	Ramechaap, Bhaktapur	Diarrhea, Menstrual disorder, Blood sugar control (Pradhan et al., 2020; Dulal et al., 2022)
336	*Momordica charantia* L. Cucurbitaceae	Bhaktapur	Blood pressure and sugar control (Dulal et al., 2022)
337	*Morina longifolia* Wall. ex DC. Caprifoliaceae	Kavrepalanchowk	Dysentery, Diarrhoea, Edible stem, Incense (Malla & Chettri , 2009)
338	*Morus alba* L. Moraceae	Nuwakot	Fodder, Edible fruits (Nepali et al. 2020)
339	*Morus macroura* Miq. Moraceae	Kavrepalanchowk	Cuts, Wounds, Fever, Sore throat, Dyspepsia, Melancholia, Edible fruits, Fodder (Malla & Chettri , 2009)
340	*Morus serrata* Roxb. Moraceae	Dolakha	Fracture (Karki et al., 2023)
341	*Mucuna interrupta* Gagnep. Fabaceae	Sinduli	Cuts, Wounds (Kafle & Dani, 2022)
342	*Murraya koenigii* (L.) Spreng. Rutaceae	Ramechaap Makawanpur	Poisonous animal bites, Dysentery, Skin diseases, Vomiting, Diarrhea, Inflammations, Flavoring, Edible fruits (Pradhan et al., 2020; Moktan, 2023)
343	*Musa* × *paradisiaca* L. Musaceae	Ramechaap, Kavrepalanchowk, Makawanpur, Chitwan	Chest pain, Dysentery, Colic disease, Intestinal disorders, Diabetes, Uremia, Nephritis, Gout, Hypertension, Cardiac disease, Diarrhea, Joint pain, Body heating, Excess bleeding, Edible flowers and fruits, Pickles, Vegetable, Plates, Religious purposes (Malla & Chhetri, 2009; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Moktan, 2023)
344	*Musa balbisiana* Colla Musaceae	Sinduli	Asthma, Fish bone/Bone pricking (Kafle & Dani, 2022)
345	*Myrica esculenta* Buch.-Ham. ex D.Don Myricaceae	Chitwan, Ramechaap, Makawanpur, Rasuwa, Dhading, Sindupalchowk, Bhaktapur, Kathmandu	Abdominal pain, Sinusitis, Gastritis, Bone fracture, Bleeding control, Toothache, Diarrhea, Dysentery, Cholera, Piles, Fever, Headache, Heart disease, Edible fruits (Balami, 2004; Rai et al., 2004; Shrestha et al., 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Gurung, 2023; Moktan, 2023)
346	*Myristica fragrans* Houtt. Myristicaceae	Lalitpur, Sinduli	Pneumonia, Skin rashes (Panta, 2019; Kafle & Dani, 2022)
347	*Myrsine capitellata* Wall. Myrsinaceae	Makawanpur	Fodder (Moktan, 2023)
348	*Nardostachys jatamansi* (D.Don) DC. Caprifoliaceae	Bhaktapur, Rasuwa	Mental weakness, Insomnia, Epilepsy, Incense (Shrestha et al., 2014; Bhaila et al., 2022)
349	*Nasturtium officinale* R.Br.	Bhaktapur	Blood sugar and pressure control (Dulal et al., 2022)

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	Brassicaceae		
350	*Neolamarckia cadamba* (Roxb.) Bosser Rubiaceae	Makawanpur	Wounds between toes, Diarrhea, Dysentery, Cuts, Wounds (Joshi et al., 2020)
351	*Neopicrorhiza scrophulariiflora* (Pennell) D.Y.Hong Scrophulariaceae	Rasuwa, Ramechaap, Sinduli	Stomach problems, Fever, Headache, Stomachache, Lactation (Shah & Singh, 2014; Shrestha et al., 2014; Pradhan et al., 2020)
352	*Nephrolepis cordifolia* (L.) C.Presl Nephrolepidaceae	Makawanpur, Nuwakot, Dolakha, Ramechaap, Lalitpur, Bhaktapur	Dehydration, Appetite stimulation, Cooling tonic, Blood sugar, Gastritis, Jaundice, Boils, Skin problems, Liver ailments, Indigestion, Headache, Cough, Fever, Uteritis in buffalo (Timilsina & Singh, 2015; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
353	*Nerium oleander* L. Apocynaceae	Sinduli	Cuts, Wounds, Bleeding control in women (Kafle & Dani, 2022)
354	*Nicotiana tabacum* L. Solanaceae	Bhaktapur, Makawanpur, Lalitpur, Sinduli, Dolakha	Sedative, Narcotic, Antiseptic, Antidote for scorpion/lice bites, Burns, Leech removal, Bruises and wounds in cattle, Insecticide, Smoking (Bhattarai & Tamang, 2017; Dhital et al., 2021; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Moktan, 2023)
355	*Nyctanthes arbor-tristis* L. Oleaceae	Dolakha, Nuwakot, Chitwan, Makawanpur, Sinduli, Dhading, Bhaktapur, Lalitpur, Kathmandu, Sindupalchowk	Asthma, Cough, Blisters, Wounds, Blood sugar control, Fever, Blood pressure, Heat, Jaundice, Piles, Fracture, Rheumatic disorder, Uric acid (Balami, 2004; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Gurung & Subedi, 2021; Bhaila et al., 2022; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Gurung, 2023; Karki et al., 2023)
356	*Ocimum basilicum* L. Lamiaceae	Chitwan, Lalitpur, Dhading, Ramechaap, Sinduli, Makawanpur	Fever, Cough, Cold, Headache, Toothache, Low blood pressure, Stomach problems, Jaundice, Pickles, Pesticide (Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Pradhan et al., 2020; Gurung & Subedi, 2021; Kafle & Dani, 2022)
357	*Ocimum gratissimum* L. Lamiaceae	Makawanpur	Edible seeds (Moktan, 2023)
358	*Ocimum tenuiflorum* L. Lamiaceae	Nuwakot, Chitwan, Sinduli, Lalitpur, Ramechaap, Bhaktapur, Dolakha, Sindupalchowk, Makawanpur	Fever, Jaundice, Cough, Cold, Typhoid, Skin disease, Anorexia, Bronchitis, Dysentery, Diarrhea, Tonsillitis, Throat pain, Toothache, Religious worship (Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Pradhan et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023; Karki et al., 2023)
359	*Onoclea struthiopteris* Roth Onocleaceae	Chitwan	Diarrhoea, Blood in stool (Poudel & Singh, 2016)
360	*Ophiocordyceps sinensis* (Berk.) G.H.Sung, J.M.Sung, Hywel-Jones & Spatafora Ophiocordycipitaceae	Ramechaap, Rasuwa	Body strength enhancement, Additives with milk/honey, Commercial value (Shrestha et al., 2014; Pradhan et al., 2020)
361	*Ophioglossum reticulatum* L. Ophioglossaceae	Sinduli	Dysentery, Diarrhea (Kafle & Dani, 2022)
362	*Opuntia monacantha* (Willd.) Haw. Euphorbiaceae	Sinduli	Fever, Cooling agent (Kafle & Dani, 2022)
363	*Origanum majorana* L. Lamiaceae	Makawanpur	Cold relief through aroma (Moktan, 2023)
364	*Oroxylum indicum* (L.) Kurz Bignoniaceae	Makawanpur, Dolakha, Dhading, Sinduli, Nuwakot, Bhaktapur	Fracture, Jaundice, Blood in stool, Appetite loss, Pneumonia, Rheumatic problems, Burns, Pimples, Boils, Fever, Wounds, Religious purposes (Bhattarai & Tamang, 2017; Joshi et al., 2020; Nepali et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Karki et al., 2023)
365	*Oryza sativa* L. Poaceae	Kavrepalanchowk, Sinduli	Scorpion bite, Heart inflammation, Indigestion, Animal feed, Mat making, Shoes, Food, Alcoholic beverage (Malla & Chhetri, 2009; Kafle & Dani, 2022)
366	*Osbeckia nepalensis* Hook. Melastomataceae	Dolakha, Kathmandu	Cuts, Wounds (Balami, 2004; Karki et al., 2023)
367	*Osbeckia nutans* Wall. ex C.B.Clarke Melastomataceae	Makawanpur	Stomach ache (Joshi et al., 2020)
368	*Osbeckia stellata* Buch.-Ham. ex D.Don Melastomataceae	Makawanpur, Kathmandu	Scabies (Balami, 2004; Joshi et al., 2020)
369	*Osyris lanceolata* Hochst. & Steud. Santalaceae	Lalitpur, Bhaktapur, Dolakha	Fracture, Hair growth promotion (Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
370	*Ototropis conferta* (DC.) H.Ohashi & K.Ohashi Fabaceae	Makawanpur	Diarrhea, Dysentery, Fever, Gastric (Joshi et al., 2020; Moktan, 2023)
371	*Ougeinia oojeinensis* (Roxb.) Hochr. Fabaceae	Makawanpur	Fodder, Dysentery (Moktan, 2023)
372	*Oxalis corniculata* L. Oxalidaceae	Chitwan, Lalitpur, Dhading, Ramechaap, Makawanpur, Dolakha, Sinduli, Kathmandu	Wounds, Eye problems, Tooth corrosion, Earache, Snake bite, Stomach problems, Joint pain, Dysentery, Fever, Diarrhea, Abdominal disorder, Liver cirrhosis, Skin diseases, Ringworm (Balami, 2004; Dangol & Gurung, 2008; Poudel & Singh, 2016; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Gurung & Subedi, 2021; Kafle & Dani, 2022; Karki et al., 2023)
373	*Paederia foetida* L. Rubiaceae	Makawanpur, Sinduli	Toothache, Appetite stimulation (Kafle & Dani, 2022; Moktan, 2023)
374	*Paris polyphylla* Sm. Melanthiaceae	Makawanpur, Dolakha, Rasuwa, Ramechaap,	Burns, Diarrhea, Dysentery, Appetite stimulation, Toothache, Fever, Antiseptic, Tonic, Anthelmintic, Gastritis, Bone fracture, Stomach disorders, Cold, Poison, Veterinary uses (Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
375	*Parmelia nepaulensis* Parmeliaceae	Ramechaap	Wounds, Cuts (Pradhan et al., 2020)
376	*Perilla frutescens* (L.) Britton Lamiaceae	Makawanpur	Edible seeds for pickling (Moktan, 2023)
377	*Persea americana* Mill. Lauraceae	Bhaktapur	Cholesterol reduction (Dulal et al., 2022)
378	*Persicaria barbata* (L.) Hara Polygonaceae	Chitwan	Swelling reduction (Dangol & Gurung, 2008)
379	*Persicaria hydropiper* (L.) Delarbre Polygonaceae	Makawanpur	Worm treatment for cattle (Moktan, 2023)
380	*Phalaris arundinacea* L. Poaceae	Bhaktapur	Albinism (Dulal et al., 2022)
381	*Phanera scandens* (L.) Lour. ex Raf. Fabaceae	Kavrepalanchowk	Stomach disorder, Skin diseases, Indigestion, Diarrhoea, Headache, Fodder, Fuel, Construction (Malla & Chettri , 2009)
382	*Phanera vahlii* (Wight & Arn.) Benth. Fabaceae	Ramechaap Makawanpur, Nuwakot	Bleeding, Dysentery, Diarrhea, Indigestion, Toxicity, Fodder, Rope making, Death rituals (Nepali et al., 2020; Pradhan et al., 2020; Moktan, 2023)

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383	*Phaseolus vulgaris* L. Fabaceae	Dhading	Ringworm (Gurung & Subedi , 2021)
384	*Phoenix loureiroi* Kunth Arecaceae	Makawanpur, Dhading, Sinduli	Itching, Pneumonia, Urinary disorder, Edible fruits and roots, Thatching, Door making (Joshi et al., 2020; Kafle & Dani, 2022; Moktan, 2023)
385	*Phoenix sylvestris* (L.) Roxb. Arecaceae	Sinduli	Body pain/Relaxation (Kafle & Dani, 2022)
386	*Phyla nodiflora* (L.) Greene Verbenaceae	Chitwan	Plant juice for stomachache, ulcers. (Dangol & Gurung, 2008)
387	*Phyllanthus emblica* L. Phyllanthaceae	Makawanpur, Dolakha, Nuwakot, Dhading, Sinduli, Bhaktapur, Ramechaap, Sindupalchowk, Chitwan, Lalitpur, Kathmandu	Bark, leaves, fruit juice for gastritis, dysentery, constipation, cold/cough, weakness, blood purification, diarrhea, urination issues, hemorrhage, scurvy, snakebite, vitamin source, antidandruff, wounds, fracture. Fruits edible, leaves used as fodder. (Balami, 2004; Rai et al., 2004; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Gurung, 2023; Karki et al., 2023; Moktan, 2023)
388	*Phyllanthus parvifolius* Buch.-Ham. ex D.Don Phyllanthaceae	Bhaktapur, Makawanpur, Dolakha	Bone fracture, cuts, dislocation.(Joshi et al., 2020; Dulal et al., 2022; Karki et al., 2023)
389	*Phyllanthus urinaria* L. Phyllanthaceae	Kavrepalanchowk	Tuber juice for diarrhea, dysentery. Leaf paste for gonorrhea, urinary troubles, boils. Ornamental. (Malla & Chettri , 2009)
390	*Phyllanthus velutinus* (Wight) Müll.Arg. Phyllanthaceae	Makawanpur	Pimples (Joshi et al., 2020)
391	*Physalis minima* L. Solanaceae	Chitwan	Leaf juice for menstrual disorders, eye infection. (Dangol & Gurung, 2008)
392	*Phytolacca acinosa* Roxb. Phytolaccaceae	Bhaktapur, Makawanpur	Eyesight, vitamin, infertility.(Joshi et al., 2020; Bhaila et al., 2022)
393	*Phytolacca americana* L. Phytolaccaceae	Bhaktapur	Blisters, jaundice, sinusitis. (Bhaila et al., 2022)
394	*Picris hieracioides* L. Asteraceae	Makawanpur	Headache (Joshi et al., 2020)
395	*Pieris formosa* (Wall.) D.Don Ericaceae	Makawanpur, Rasuwa	Leaf/stem extract for intestinal worms, scabies.(Shrestha et al., 2014; Joshi et al., 2020)
396	*Pinus roxburghii* Sarg. Pinaceae	Dolakha, Nuwakot, Ramechaap	Resin, seed, bark for indigestion, cough/cold, skin disease, asthma, tuberculosis. Timber, furniture, construction. Incense. (Nepali et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
397	*Pinus wallichiana* (Wall. ex. D.Don) A.B.Jacks. Pinaceae	Ramechaap	Timber. Resin for cuts, wounds. Incense. (Pradhan et al., 2020)
398	*Piper betle* L. Piperaceae	Sinduli	Anorexia, tonsillitis. (Shah & Singh, 2014)
399	*Piper longum* L. Piperaceae	Chitwan, Makawanpur, Ramechaap, Sinduli,	Fruit juice for fever, cough, cold, urinary infections.(Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Kafle & Dani, 2022)
400	*Piper nigrum* L. Piperaceae	Bhaktapur, Lalitpur, Sinduli	Pus, fever, stomach pain, gastritis. (Panta, 2019; Bhaila et al., 2022; Dulal et al., 2022; Kafle & Dani, 2022)
401	*Plantago asiatica* subsp. *erosa* (Wall.) Z.Yu Li Plantaginaceae	Makawanpur, Dolakha	Anthelmintic. Root juice for snake bite. (Joshi et al., 2020; Karki et al., 2023)
402	*Plantago major* L. Plantaginaceae	Ramechaap, Bhaktapur	Anthelmintic. Plant juice for fever, stomach problems, cough, cold. (Pradhan et al., 2020)
403	*Plumbago zeylanica* L. Plumbaginaceae	Sindupalchowk, Ramechaap, Sinduli, Bhaktapur, Makawanpur	Root juice/paste for skin disease, fungal infection, appetite, abdominal disorders, cold/cough, asthma, constipation, diarrhea, dysentery, scabies.(Rai et al., 2004; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022)
404	*Plumeria rubra* L. Apocynaceae	Dolakha	Latex for toothache. (Karki et al., 2023)
405	*Podophyllum hexandrum* Royle Berberidaceae	Makawanpur	Wound infection (Joshi et al., 2020)
406	*Pogostemon benghalensis* Kuntze Lamiaceae	Nuwakot, Sinduli, Makawanpur, Bhaktapur	Leaves paste/juice for antidandruff, antifungal, skin disease, warts, wounds, cough, cold, toothache, fever, sinusitis, typhoid, diarrhea, vomiting. (Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Joshi et al., 2020; Bhaila et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Moktan, 2023)
407	*Pogostemon parviflorus* Benth. Lamiaceae	Chitwan	Fresh juice after three days of fever. (Dangol & Gurung, 2008)
408	*Potentilla indica* (Andrews) Th.Wolf Rosaceae	Chitwan, Sindupalchowk	Lice infestation, fever, urinary disorders.(Dangol & Gurung, 2008; Gurung, 2023)
409	*Potentilla peduncularis* D.Don Rosaceae	Rasuwa	Root paste with water for headache. (Shrestha et al, 2014)
410	*Pouzolzia rugulosa* (Wedd.) Acharya & Kravtsova Urticacae	Kavrepalanchowk	Bark juice for cuts, wounds, body pain. Bark powder for soft bread. Wood for household materials. Fodder. (Malla & Chettri , 2009)
411	*Pouzolzia sanguinea* (Blume) Merr. Urticacae	Dolakha	Urination blockade. (Dhital et al., 2021)
412	*Pouzolzia zeylanica* (L.) Benn. Urticacae	Chitwan	Plant juice for ringworm infections, wounds on fingers and toes. (Dangol & Gurung, 2008)
413	*Premna barbata* Wall. Verbenaceae	Chitwan Makawanpur, Sinduli	Fever, cuts, wounds, thirst. (Dangol & Gurung, 2008; Joshi et al., 2020; Kafle & Dani, 2022)
414	*Premna serratifolia* L. Lamiaceae	Makawanpur, Sinduli	Typhoid (Joshi et al., 2020; Kafle & Dani, 2022)
415	*Prunus armeniaca* L. Rosaceae	Bhaktapur	Body hotness. (Bhaila et al., 2022)
416	*Prunus cerasoides* D.Don Rosaceae	Ramechaap, Dolakha, Nuwakot	Bark paste for broken body parts, intestinal worms, tapeworm. Fruits edible. Religious purposes. Fodder.(Nepali et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
417	*Prunus domestica* L. Rosaceae	Dolakha	Bark paste for fracture. (Karki et al., 2023)
418	*Prunus persica* (L.) Batsch Rosaceae	Makawanpur, Sinduli, Nuwakot, Lalitpur	Anthelmintic, bronchitis. Leaf paste for animal hoof suppuration, wounds, foot & mouth disease.(Shah & Singh, 2014; Bhattarai & Tamang, 2017; Nepali et al., 2020; Nagarkoti & Shrestha, 2022; Moktan, 2023)

S. N.	Species/Family	Reported districts	Uses
419	*Psidium guajava* L. Myrtaceae	Chitwan, Kavrepalanchowk, Lalitpur, Ramechaap Makawanpur, Sidhuli, Nuwakot, Bhaktapur	For diarrhea, dysentery, fever, headache, bowels, wounds, ulcers, cholera, indigestion, rheumatism, sugar control, bleeding gum. Fruits edible. Toothbrush. Fodder. (Malla & Chhetri, 2009; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Moktan, 2023)
420	*Pterocarpus santalinus* L.f. Fabaceae	Ramechaap	Bark/heartwood paste/powder for stomach problems, skin disease, fever, mental issues. Religious purposes. Timber. (Pradhan et al., 2020)
421	*Punica granatum* L. Lythraceae	Ramechaap Nuwakot	Bark paste for inflammation. Fruit rind powder for diarrhea, dysentery. Bark as antifungal, wound healing. For low blood pressure. Fruits edible. (Timilsina & Singh, 2015; Pradhan et al., 2020)
422	*Pyracantha crenulata* (D.Don) M.Roem. Rosaceae	Dolakha	Small branches for cattle during delivery to remove placenta. (Karki et al., 2023)
423	*Pyrostegia venusta* (Ker Gawl.) Miers Bignoniaceae	Bhaktapur	Diarrhea, dysentery. (Bhaila et al., 2022)
424	*Pyrularia edulis* (Wall.) A.DC. Cervantesiaceae	Dolakha	Seed oil for skin dryness, burns, scabies, animal neck dryness. (Karki et al., 2023)
425	*Pyrus pashia* Buch.-Ham. ex D.Don Rosaceae	Dolakha, Nuwakot	Fruit juice for animal eye problems, wounds, eye strain, excessive menstrual cycle. (Karki et al., 2023; Timilsina & Singh, 2015)
426	*Quercus lanata* Sm. Fagaceae	Bhaktapur, Dolakha, Ramechaap, Sindupalchowk	Bark paste for body pain, fracture, gastritis, pyorrhea. Fodder. Heartwood for agricultural equipment. (Rai et al., 2004; Pradhan et al., 2020; Bhaila et al., 2022; Karki et al., 2023)
427	*Quercus semecarpifolia* Sm. Fagaceae	Nuwakot	Timber, carpentry, firewood, fodder. (Nepali et al. 2020)
428	*Ranunculus sceleratus* L. Ranunculaceae	Bhaktapur	Appetizer (Dulal et al., 2022)
429	*Rauvolfia serpentina* Benth. ex Kurz Apocynaceae	Chitwan, Sinduli, Makawanpur	Snakebite, fever, urinary problem, wounds. (Dangol & Gurung, 2008; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017)
430	*Reinwardtia indica* Dumort. Linaceae	Bhaktapur, Sindupalchowk	Headache, jaundice. (Rai et al., 2004; Bhaila et al., 2022)
431	*Rhaphidophora decursiva* (Roxb.) Schott Araceae	Dolakha	Stem paste for cuts, healing wounds. (Karki et al., 2023)
432	*Rheum acuminatum* Hook.f. & Thomson Polygonaceae	Ramechaap	Root juice for fever, cough/cold, body pain. Petioles for pickles. May cause gastritis in excess. (Pradhan et al., 2020)
433	*Rheum australe* D.Don Polygonaceae	Dolakha, Rasuwa, Makawanpur	Root paste for diarrhea, sprain, body pain. Leaves/stems for pickles. (Shrestha et al., 2014; Bhattarai & Tamang, 2017; Dhital et al., 2021)
434	*Rheum spiciforme* Royle Polygonaceae	Rasuwa	Root paste for diarrhea. Leaves/stems for pickles. (Shrestha et al, 2014)
435	*Rhododendron arboreum* Sm. Ericaceae	Dhading, Rasuwa, Chitwan, Lalitpur, Ramechaap, Dolakha, Sinduli, Nuwakot, Bhaktapur, Sindupalchowk	For bloody dysentery, giardiasis, diarrhea, cough/colds, wounds, bleeding, throat pain, neck issues, stone problems, voice clarity. Dry petals for bone splinter extraction. Firewood, neck pain medicine. Pickled flowers. (Rai et al., 2004; Shrestha et al., 2014; Poudel & Singh, 2016; Pradhan et al., 2020; Gurung & Subedi, 2021; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023)
436	*Rhododendron lepidotum* Wall. ex G.Don Ericaceae	Makawanpur	Dysentery (Joshi et al., 2020)
437	*Rhododendron setosum* D.Don Ericaceae	Ramechaap, Rasuwa	Leaf juice/paste as tonic, for fever, cough/cold, joint/body pain, headaches, gastritis, stomach disorders, vein pain. Plant odor as anti-poison for animals. Essential oil, perfumes. Incense. (Shrestha et al., 2014; Pradhan et al., 2020)
438	*Rhus chinensis* Mill. Anacardiaceae	Dhading, Kavrepalanchowk, Sindupalchowk	Ground fruits for paralysis, colic, gastritis, diarrhea, bloody dysentery, swellings, wounds. Chewed fruits for stomach issues, appetite. Fruit powder for menstruation. Fruit decoction for animal foot/mouth diseases. Fruits eaten fresh/pickled.(Malla & Chhetri, 2009; Gurung & Subedi, 2021; Gurung, 2023)
439	*Rhynchostylis retusa* (L.) Blume Orchidaceae	Dolakha	Leaf paste for cuts. (Karki et al., 2023)
440	*Ricinus communis* L. Euphorbiaceae	Sinduli, Makawanpur, Kathmandu	Amoebic dysentery, foot/hand cracks & fissures. (Balami, 2004; Bhattarai & Tamang, 2017; Kafle & Dani, 2022)
441	*Rohdea nepalensis* (Raf.) N.Tanaka Asparagaceae	Makawanpur	Wound with pus. (Joshi et al., 2020)
442	*Roscoea auriculata* K.Schum. Zingiberaceae	Ramechaap	Root juice/powder as tonic, for urinary problems, fever, body strength. Root edible. (Pradhan et al., 2020)
443	*Rubia manjith* Roxb. Rubiaceae	Ramechaap, Makawanpur, Dolakha, Rasuwa, Kathmandu	Whole plant for dye. Root/stem paste/juice for diarrhea, dysentery, wound, jaundice, broken bones, diabetes, cough/cold, skin diseases, insect bite. (Balami, 2004; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
444	*Rubus ellipticus* Sm. Rosaceae	Sindupalchowk, Ramechaap, Makawanpur, Dolakha, Nuwakot, Lalitpur, Rasuwa, Bhaktapur, Kathmandu	For typhoid, fever, measles, cold, gastric, dysentery, snake bite, diarrhea, indigestion, throat pain, bone fracture, pneumonia, headache, stomachache, wounds, neck ache, jaundice. Tonic, toothpaste. Fruit edible. (Balami, 2004; Rai et al., 2004; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Nepali et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023; Karki et al., 2023; Moktan, 2023)
445	*Rumex nepalensis* Spreng. Polygonaceae	Ramechaap, Makawanpur, Dolakha, Rasuwa, Lalitpur	Boiled leaf juice for lung disease, ringworm, itching, cough/cold, sinusitis, headache, wound, cracked heels/cheeks, fracture, sprain, cut bleeding, stomachache, body pain, jaundice, swelling. Tender leaves/shoots as vegetables.(Shrestha et al., 2014; Joshi et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
446	*Rungia pectinata* (L.) Nees Acanthaceae	Chitwan,	Cuts and wounds. (Poudel & Singh, 2016)
447	*Saccharum officinarum* L. Poaceae	Sinduli, Kavrepalanchowk, Bhaktapur, Lalitpur, Sindupalchowk, Chitwan	For jaundice, anemia, heat, urinary problems, stomach disorders, skin ulcers, seminal weakness. Tonic, blood purifier. Leaves for animal placenta expulsion. Ceremonial/religious uses. (Rai et al., 2004; Malla & Chhetri, 2009; Shah & Singh, 2014; Poudel & Singh, 2016; Dulal et al., 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023)
448	*Saccharum spontaneum* L. Poaceae	Makawanpur	Thatching, fodder (Moktan, 2023)
449	*Santalum album* L. Santalaceae	Ramechaap, Makawanpur	Heartwood paste for skin problems, joint pain, uterus prolapse, body cooling. Religious purposes. Timber.(Pradhan et al., 2020; Gautam & Dhakal, 2023)
450	*Sapindus mukorossi* Gaertn. Sapindaceae	Lalitpur, Ramechaap, Nuwakot, Bhaktapur, Makawanpur	Fruit lather for burns, pregnancy, hair problems, joining nerves. Timber, fuelwood. Soap, insecticide.(Bhattarai & Tamang, 2017; Panta, 2019; Nepali et al., 2020; Pradhan et al., 2020; Dulal et al., 2022)
451	*Sarcococca coriacea* Sweet Buxaceae	Makawanpur, Kathmandu	Fever(Balami, 2004; Joshi et al., 2020)
452	*Sarcococca wallichii* Stapf Buxaceae	Dolakha	Poisonous root juice for intestinal worms. (Karki et al., 2023)
453	*Satyrium nepalense* D.Don	Makawanpur, Kathmandu	(Balami, 2004; Joshi et al., 2020)

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	Orchidaceae		
454	*Saurauia napaulensis* DC. Actinidiaceae	Dolakha, Makawanpur	Pneumonia, fruit jelly for cough. (Bhattarai & Tamang, 2017; Karki et al., 2023)
455	*Saussurea gossypiphora* Franch. Asteraceae	Rasuwa	Root extract for fever. (Shrestha et al, 2014)
456	*Schima wallichii* (DC.) Korth. Theaceae	Bhaktapur, Dhading, Ramechaap, Makawanpur, Dolakha, Nuwakot, Sinduli, Sindupalchowk, Kathmandu	Bark paste for cuts, wounds, swollen areas, bone fracture, gastric, skin rashes, taeniasis. Bark powder for animal ailments. Timber, firewood, manure. (Balami, 2004; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Bhaila et al., 2022; Kafle & Dani, 2022; Gurung, 2023; Karki et al., 2023)
457	*Schleichera oleosa* (Lour.) Oken Sapindaceae	Makawanpur	Ripe fruit pulp edible. (Moktan, 2023)
458	*Scindapsus officinalis* (Roxb.) Schott Araceae	Dolakha	Bone fracture. (Dhital et al., 2021)
459	*Scoparia dulcis* L. Plantaginaceae	Chitwan, Makawanpur, Sinduli	Root paste for typhoid. Leaf juice for fever cooling, throat sore. (Dangol & Gurung, 2008; Joshi et al., 2020; Kafle & Dani, 2022; Moktan, 2023)
460	*Scurrula parasitica* L. Loranthaceae	Makawanpur	Bone fracture. (Joshi et al., 2020)
461	*Scutellaria discolor* Wall. ex Benth. Lamiaceae	Dhading, Makawanpur, Kathmandu	Fever (Balami, 2004; Bhattarai & Tamang, 2017; Joshi et al., 2020; Gurung & Subedi, 2021; Moktan, 2023)
462	*Semecarpus anacardium* L.f. Anacardiaceae	Makawanpur, Bhaktapur	Anthelmintic, fever, latex-induced skin irritation. (Bhattarai & Tamang, 2017; Joshi et al., 2020; Dulal et al., 2022)
463	*Senegalia catechu* (L.f.) P.J.H.Hurter & Mabb. Fabaceae	Sinduli	Cough (Kafle & Dani, 2022)
464	*Senegalia pennata* (L.) Maslin Fabaceae	Sinduli	Kidney stone. (Kafle & Dani, 2022)
465	*Senna occidentalis* (L.) Link Fabaceae	Chitwan	Root bark paste for ringworm infection. (Dangol & Gurung, 2008)
466	*Senna tora* (L.) Roxb. Fabaceae	Makawanpur Sinduli	Fever, rheumatism, anthelmintic, itching. (Joshi et al., 2020; Kafle & Dani, 2022)
467	*Sesamum indicum* L. Pedaliaceae	Sinduli Ramechaap	Seed/root juice/paste for dysentery, bleeding, constipation, skin dryness, malaria. Oil extraction. Seeds eaten as pickles or directly. (Pradhan et al., 2020; Kafle & Dani, 2022)
468	*Shorea robusta* C.F.Gaertn. Dipterocarpaceae	Chitwan, Bhaktapur, Sinduli, Makawanpur, Nuwakot	Bark juice for diarrhea, dysentery, cough, rheumatism, fracture, piles, cancer, stone, herpes zoster. Timber, fodder, leaf plates. (Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Nepali et al., 2020; Bhaila et al., 2022; Kafle & Dani, 2022; Moktan, 2023)
469	*Sida acuta* Burm.f. Malvaceae	Makawanpur	Boils (Joshi et al., 2020)
470	*Sida cordifolia* L. Asteraceae	Chitwan, Makawanpur	Leaf/stem juice for boil pus extraction. (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017)
471	*Smallanthus sonchifolius* (Poepp.) H.Rob. Asteraceae	Bhaktapur	Anti-diabetes. (Dulal et al., 2022)
472	*Smilax aspera* L. Smilacaceae	Ramechaap, Dolakha, Nuwakot, Sindupalchowk, Makawanpur, Kathmandu	Root paste for syphilis, skin disease, venereal diseases, delivery disorder. Young shoots for cancer. Religious purposes. Making dhyangro, fodder. (Balami, 2004; Rai et al., 2004; Bhattarai & Tamang, 2017; Nepali et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
473	*Smilax ovalifolia* Roxb. Smilacaceae	Makawanpur, Dhading	Eyesight tonic, menstrual disorder.(Joshi et al., 2020; Gurung & Subedi, 2021)
474	*Smilax purhampuy* Ruiz Smilacaceae	Dolakha	Warding evil spirit, abortion. (Dhital et al., 2021)
475	*Solanum aculeatissimum* Jacq. Solanaceae	Chitwan, Makawanpur, Sinduli, Kathmandu	Fruit smoke for toothache, fever, headache.(Balami, 2004; Dangol & Gurung, 2008; Joshi et al., 2020; Kafle & Dani, 2022)
476	*Solanum capsicoides* All. Solanaceae	Chitwan	Asthma, chest pain. (Poudel & Singh, 2016)
477	*Solanum lycopersicum* L. Solanaceae	Bhaktapur	Fever, gastric, cough. (Dulal et al., 2022)
478	*Solanum melongena* L. Solanaceae	Kavrepalanchowk, Chitwan	Root decoction for heart problems. Leaf juice for throat aches, stomach problems. Roasted green fruit for cough/cold, placenta retention. Fruits cooked as vegetable, pickled. (Malla & Chhetri, 2009; Poudel & Singh, 2016)
479	*Solanum pseudocapsicum* L. Solanaceae	Bhaktapur, Kathmandu	Piles (Balami, 2004; Dulal et al., 2022)
480	*Solanum virginianum* L. Solanaceae	Bhaktapur, Nuwakot, Makawanpur	Toothache, sore throat. (Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Dulal et al., 2022)
481	*Solena amplexicaulis* (Lam.) Gandhi Cucurbitaceae	Bhaktapur, Makawanpur, Dolakha	For dysentery, fever, tonic, indigestion, throat pain, stomatitis, stone problem, appetite, typhoid, urine inflammation. Fruit for fever, root juice for neck pain. (Joshi et al., 2020; Bhaila et al., 2022; Karki et al., 2023)
482	*Sonchus oleraceus* L. Asteraceae	Bhaktapur	Earache, cut and wound. (Dulal et al., 2022)
483	*Sonchus wightianus* DC. Asteraceae	Makawanpur	Diabetes. Leaves/shoots as vegetable. Leaves on abdomen for easier delivery.(Joshi et al., 2020; Moktan, 2023)
484	*Spermadictyon suaveolens* Roxb. Rubiaceae	Makawanpur	Cuts and wounds. (Joshi et al., 2020)
485	*Spilanthes paniculata* DC. Asteraceae	Makawanpur	Plant paste for fish poisoning.(Bhattarai & Tamang, 2017; Moktan, 2023)
486	*Spondias pinnata* (L.f.) Kurz Anacardiaceae	Nuwakot, Sinduli	Common cold/cough. Firewood, pickle. (Nepali et al., 2020; Kafle & Dani, 2022)
487	*Stellaria media* (L.) Vill. Caryophyllaceae	Dolakha	Cuts, wounds. (Dhital et al., 2021)
488	*Stellaria monosperma* Buch.-Ham. ex D.Don Caryophyllaceae	Makawanpur	Diarrhea (Bhattarai & Tamang, 2017; Joshi et al., 2020)
489	*Stephania glandulifera* Miers Menispermaceae	Ramechaap, Makawanpur, Dolakha	Root juice for constipation, bleeding, skin disease, gastritis, diarrhea, indigestion, menstrual disorder. Tuberous roots for cattle diarrhea. (Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
490	*Sterculia villosa* Roxb.	Makawanpur	Bark for ropes. Powdered root with rice flour for bread. Fruits edible. (Moktan, 2023)

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	Malvaceae		
491	*Stereospermum chelonoides* (L.f.) DC. Bignoniaceae	Chitwan	Dried stem bark powder for leprosy. (Dangol & Gurung, 2008)
492	*Strobilanthes lachenensis* C.B.Clarke Acanthaceae	Dolakha	Rhizome chewed for sore throat, cough/cold. Rhizome juice for scabies, skin diseases. (Karki et al., 2023)
493	*Swertia angustifolia* Buch.-Ham. ex D.Don Gentianaceae	Kavrepalanchowk, Makawanpur, Rasuwa, Kathmandu	Plant extract for fever. Plant decoction as blood purifier, for bile disease, cough/cold. (Balami, 2004; Malla & Chhetri, 2009; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Moktan, 2023)
494	*Swertia chirayita* (Roxb.) H.Karst. Gentianaceae	Ramechaap, Makawanpur, Rasuwa, Sindupalchowk, Dolakha, Kathmandu	Cough, fever, appetite, blood pressure, headache, body pain, anxiety.(Balami, 2004; Rai et al., 2004; Shrestha et al., 2014; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Karki et al., 2023)
495	*Swertia multicaulis* D.Don Gentianaceae	Rasuwa	Root paste for bleeding prevention, wound/cut infection, burns. Roots as buti for warding evil spirits. (Shrestha et al, 2014)
496	*Swertia nervosa* (G.Don) Wall. ex C.B.Clarke Gentianaceae	Chitwan, Kathmandu	Cuts and wounds. (Balami, 2004; Poudel & Singh, 2016)
497	*Symplocos pyrifolia* Wall. & G.Don Symplocaceae	Ramechaap Makawanpur Dolakha	Seeds for edible oil. Edible fruits. Seed paste for skin diseases, dryness. (Pradhan et al., 2020; Karki et al., 2023; Moktan, 2023)
498	*Syzygium aromaticum* (L.) Merr. & L.M.Perry Myrtaceae	Chitwan, Sinduli	Tonsillitis, toothache. (Poudel & Singh, 2016; Kafle & Dani, 2022)
499	*Syzygium cumini* (L.) Skeels Myrtaceae	Bhaktapur, Ramechaap, Makawanpur, Sinduli, Kathmandu	Bark/leaf juice for diarrhea, indigestion, constipation, swollen body, diabetes. Edible fruits. Fodder. Timber, furniture, fuelwood. (Balami, 2004; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Moktan, 2023)
500	*Syzygium nervosum* A.Cunn. ex DC. Myrtaceae	Makawanpur, Dhading, Bhaktapur, Sinduli, Ramechaap	Leaf/bark powder smoke for headaches, cough, sinusitis. Timber, fuelwood. (Joshi et al., 2020; Pradhan et al., 2020; Gurung & Subedi, 2021; Bhaila et al., 2022; Kafle & Dani, 2022)
501	*Tagetes erecta* L. Asteraceae	Sinduli, Dolakha, Bhaktapur	Appetite loss, pneumonia, fever/typhoid. (Dhital et al., 2021; Dulal et al., 2022; Kafle & Dani, 2022)
502	*Tamarindus indica* L. Fabaceae	Chitwan, Sinduli, Ramechaap	Bark/root juice for diarrhea, constipation, inflammation, indigestion, rheumatic pain, scurvy. Tonic, cooling effect. Edible fruits, pickles. Fodder. Timber. (Dangol & Gurung, 2008; Shah & Singh, 2014; Pradhan et al., 2020)
503	*Taxus baccata* L. Taxaceae	Bhaktapur, Kavrepalanchowk	Leaf juice for cough/cold, bronchitis, asthma, fever, bleeding. Taxol from bark/leaves for tumors, breast/uterus cancer.(Malla & Chhetri, 2009; Bhaila et al., 2022)
504	*Taxus wallichiana* Zucc. Taxaceae	Dolakha, Rasuwa, Makawanpur	Jaundice, cancer, gastritis, asthma, bronchitis, respiratory infections (Shrestha et al., 2014; Bhattarai & Tamang, 2017; Karki et al., 2023)
505	*Tectaria coadunata* (J.Sm.) C.Chr. Tectariaceae	Nuwakot	Diarrhea and dysentery (Timilsina & Singh, 2015)
506	*Tephrosia purpurea* (L.) Pers. Fabaceae	Makawanpur	Flower cooked as vegetable. (Moktan, 2023)
507	*Terminalia bellirica* (Gaertn.) Roxb. Combretaceae	Chitwan, Sinduli, Makawanpur, Nuwakot,	For anemia, ulcers, cough, fever, skin diseases, bronchitis, constipation. Tonic, anthelmintic, diarrhea, wounds, jaundice, blood pressure, gastritis, throat pain. (Dangol & Gurung, 2008; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Nepali et al., 2020; Moktan, 2023)
508	*Terminalia chebula* Retz. Combretaceae	Chitwan, Makawanpur, Sinduli, Dolakha, Nuwakot, Bhaktapur	For cough/cold, blood pressure, sinusitis, fever, stomach, appetite, urinary problems, diarrhea, wounds, jaundice, dysentery, ulcers, neuropathy, gastritis. (Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Nepali et al., 2020; Dhital et al., 2021; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhakal, 2023; Karki et al., 2023; Moktan, 2023)
509	*Terminalia elliptica* Willd. Combretaceae	Makawanpur, Dolakha	Gastric, scabies. Timber. (Gautam & Dhakal, 2023; Karki et al., 2023)
510	*Termitomyces eurrhizus* (Berk.) R.Heim (1942) Tricholomataceae	Makawanpur	Roasted and eaten as vegetable. (Moktan, 2023)
511	*Thalictrum foliolosum* DC. Ranunculaceae	Bhaktapur, Makawanpur, Dolakha, Rasuwa	Antiseptic for ear piercing, gastritis, stomach disorder, jaundice, itching. (Joshi et al., 2020; Shrestha et al., 2014; Bhaila et al., 2022; Karki et al., 2023)
512	*Thalictrum virgatum* Hook.f. & Thomson Ranunculaceae	Sindupalchowk, Kathmandu	Sinusitis (Balami, 2004; Rai et al., 2004)
513	*Themeda triandra* Forssk. Poaceae	Makawanpur	Roofing (Moktan, 2023)
514	*Thevetia peruviana* (Pers.) K.Schum. Apocynaceae	Chitwan	Dried fruit paste for ear infection. (Dangol & Gurung, 2008)
515	*Thomsonia napalensis* Wall. Araceae	Sindupalchowk	Sexual imbalance.(Rai et al., 2004)
516	*Thysanolaena latifolia* (Roxb. ex Hornem.) Honda Poaceae	Chitwan, Ramechaap, Kavrepalanchowk, Makawanpur, Lalitpur	Root juice for boils, muscular pain, fever, constipation, body/joint pain, urinary infection, placenta retention. Fodder. Inflorescences as broom. Tender leaf base eaten fresh. (Malla & Chhetri, 2009; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Pradhan et al., 2020; Moktan, 2023)
517	*Tinospora cordifolia* (Willd.) Miers ex Hook.f. & Thomson Menispermaceae	Dolakha	Bark juice for scabies. (Karki et al., 2023)
518	*Tinospora sinensis* (Lour.) Merr. Menispermaceae	Chitwan, Bhaktapur, Ramechaap, Makawanpur, Sinduli, Sindupalchowk, Kathmandu	Plant juice for urinary tract infection, appetite, body ache, diabetes, tonic, earache, gastritis, jaundice, stomachache, fever. Stem for herbal tea. (Balami, 2004; Shah & Singh, 2014; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Joshi et al., 2020; Pradhan et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Gurung, 2023)
519	*Toona ciliata* M.Roem. Meliaceae	Nuwakot, Kavrepalanchowk, Makawanpur	Bark juice for chronic infantile dysentery, ulcers, boils, cough, bronchitis. Leaves as fodder. Wood for furniture, carving, fuel. (Malla & Chhetri, 2009; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Nepali et al., 2020)
520	*Toxicodendron wallichii* (Hook.f.) Kuntze Anacardiaceae	Makawanpur	Wounds (Joshi et al., 2020)
521	*Trachyspermum ammi* Sprague Apiaceae	Sinduli	Measles, menstrual cramps, lactation enhancer, tonic for pregnant women. (Kafle & Dani, 2022)
522	*Trametes versicolor* (L.) Lloyd Polyporaceae	Dolakha	Dysentery (Dhital et al., 2021)
523	*Tribulus terrestris* L. Zygophyllaceae	Makawanpur	Body inflammation, kidney stones, cough. (Bhattarai & Tamang, 2017; Joshi et al., 2020)
524	*Trichilia connaroides* (Wight & Arn.) Bentv. Meliaceae	Sindupalchowk	Scabies (Rai et al., 2004)
525	*Trichodesma indicum* (L.) Lehm.	Chitwan	Dried plant mixed with flour for bread to treat headache. (Dangol & Gurung, 2008)

S. N.	Species/Family	Reported districts	Uses
	Boraginaceae		
526	*Trichosanthes cucumerina* L. Cucurbitaceae	Sinduli	Jaundice (Kafle & Dani, 2022)
527	*Trichosanthes dioica* Roxb. Cucurbitaceae	Chitwan	Heat (Poudel & Singh, 2016)
528	*Trichosanthes tricuspidata* Lour. Cucurbitaceae	Makawanpur, Dolakha	Fever, liver swelling, mouth ulcer. Root paste for snakebite. (Bhattarai & Tamang, 2017; Joshi et al., 2020; Karki et al., 2023)
529	*Tridax procumbens* L. Asteraceae	Sinduli	Wound (to stop bleeding) (Kafle & Dani, 2022)
530	*Trigonella foenum-graecum* L. Fabaceae	Dolakha, Ramechaap	Seeds for appetite. Bone fracture. Leaves as vegetables.(Pradhan et al., 2020; Dhital et al., 2021)
531	*Triticum aestivum* L. Poaceae	Kavrepalanchowk	Edible grains. Roasted seeds eaten. Straw for cattle feed, craft. Grains for 'Chhang'. (Malla & Chettri , 2009)
532	*Uraria crinita* (L.) Desv. ex DC. Fabaceae	Sinduli	Blood dysentery. (Kafle & Dani, 2022)
533	*Urena lobata* L. Malvaceae	Makawanpur	Fruits eaten by goats.(Bhattarai & Tamang, 2017; Moktan, 2023)
534	*Urtica dioica* L. Urticacae	Lalitpur, Chitwan, Makawanpur, Sinduli, Ramechaap, Dolakha, Sindupalchowk, Nuwakot, Kathmandu	Root/leaf powder/paste for chest pain, gastritis, jaundice, cuts, wounds, kidney stone, toothache, blood pressure, blood sugar, body temperature. Tonic. For bone dislocation, fracture. Young leaves as vegetable. Plant juice as bath for children. For sole crack, sprain in cattle.(Balami, 2004; Rai et al., 2004; Shah & Singh, 2014; Timilsina & Singh, 2015; Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Panta, 2019; Joshi et al., 2020; Pradhan et al., 2020; Dhital et al., 2021; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gurung, 2023; Karki et al., 2023; Moktan, 2023)
535	*Valeriana jatamansi* Jones Valerianaceae	Lalitpur, Rasuwa	Root paste with water for headache, eye pain. For bathing children with weepy nature. (Shrestha et al., 2014; Nagarkoti & Shrestha, 2022)
536	*Viburnum cylindricum* Buch.-Ham. ex D.Don Viburnaceae	Dolakha	Seed oil for skin abrasions. (Karki et al., 2023)
537	*Viburnum erubescens* Wall. ex DC. Viburnaceae	Dolakha	Root paste for goiter. (Karki et al., 2023)
538	*Viburnum mullaha* Buch.-Ham. ex D.Don Viburnaceae	Dolakha	Fruits for diarrhea. (Karki et al., 2023)
539	*Vigna mungo* (L.) Hepper Fabaceae	Kavrepalanchowk	Root juice for nostalgia, abscess, inflammations. Seeds cooked as lentil soup for appetite, tonic, urinary problems. Seed flour for bread. Husk/straw as cattle fodder. (Malla & Chettri , 2009)
540	*Viola pilosa* Blume Violaceae	Makawanpur, Dolakha	Fever. Plant juice for cuts, wounds to stop bleeding.(Joshi et al., 2020; Karki et al., 2023)
541	*Viscum album* L. Santalaceae	Chitwan, Makawanpur, Dolakha	Dislocated bone, fracture. (Luitel et al., 2014; Poudel & Singh, 2016; Joshi et al., 2020; Karki et al., 2023)
542	*Vitex negundo* L. Lamiaceae	Makawanpur, Nuwakot, Sinduli, Sindupalchowk, Bhaktapur, Kathmandu	Swollen body parts, skin disease, headache, sinusitis, cough/cold. (Balami, 2004; Rai et al., 2004; Timilsina & Singh, 2015; Joshi et al., 2020; Dulal et al., 2022; Kafle & Dani, 2022; Gautam & Dhital, 2023)
543	*Withania somnifera* (L.) Dunal Solanaceae	Chitwan, Makawanpur	Weakness, abdominal disorder. (Poudel & Singh, 2016; Bhattarai & Tamang, 2017; Gautam & Dhakal, 2023)
544	*Woodfordia fruticosa* Kurz Lythraceae	Bhaktapur, Makawanpur, Dolakha, Kathmandu	Cuts and wounds, dysentery, fever, diarrhea. (Balami, 2004; Bhattarai & Tamang, 2017; Joshi et al., 2020; Bhaila et al., 2022; Karki et al., 2023; Moktan, 2023)
545	*Xanthium indicum* J.Koenig ex Roxb. Asteraceae	Makawanpur	Cuts and wounds, fracture. (Joshi et al., 2020)
546	*Xanthium strumarium* L. Asteraceae	Dolakha	Dysentery (Dhital et al., 2021)
547	*Xeromphis spinosa* (Thunb.) Keay Rubiaceae	Chitwan	Fruits as fish poison. Stem bark powder mixed with water for leprosy. (Dangol & Gurung, 2008)
548	*Xylanche himalaica* (Hook.f. & Thomson) Beck Orobanchaceae	Rasuwa	Root paste for cuts and wounds for early healing. (Shrestha et al, 2014)
549	*Zantedeschia aethiopica* (L.) Spreng. Araceae	Lalitpur	Sticky stem product for dog/snake bites.(Nagarkoti & Shrestha, 2022)
550	*Zanthoxylum acanthopodium* DC. Rutaceae	Dolakha	Fruit (Dhital et al., 2021)
551	*Zanthoxylum armatum* DC. Rutaceae	Bhaktapur, Makawanpur, Sinduli, Dolakha, Rasuwa, Nuwakot, Lalitpur, Kathmandu	For gastric/stomach issues, digestion, high blood pressure, appetite, repelling leeches, fracture, fever, cold/cough, toothache, paralysis, skin disease, ringworms. Used in curry/pickles. (Balami, 2004; Shrestha et al., 2014; Timilsina & Singh, 2015; Bhattarai & Tamang, 2017; Panta, 2019; Bhaila et al., 2022; Dulal et al., 2022; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Karki et al., 2023)
552	*Zanthoxylum bungeanum* Maxim. Rutaceae	Rasuwa	Powdered fruit for stomach disorder. Used in curry/pickles. (Shrestha et al, 2014)
553	*Zea mays* L. Poaceae	Sinduli, Kavrepalanchowk, Ramechaap	Grains for reducing sugar levels in diabetes, weakness. Edible grains. Leaves as fodder. Husks for household goods. (Malla & Chhetri, 2009; Shah & Singh, 2014; Pradhan et al., 2020)
554	*Zephyranthes carinata* Herb. Amaryllidaceae	Makawanpur	Gastric (Joshi et al., 2020)
555	*Zingiber officinale* Roscoe Zingiberaceae	Ramechaap, Sinduli, Dolakha, Lalitpur, Makawanpur, Sindupalchowk	For cough, dysentery, common cold, tonsil/throat pain, indigestion, body pain, arthritis, body warming. Rhizome eaten directly or as tea. (Luitel et al., 2014; Shah & Singh, 2014; Bhattarai & Tamang, 2017; Pradhan et al., 2020; Kafle & Dani, 2022; Nagarkoti & Shrestha, 2022; Gautam & Dhakal, 2023; Gurung, 2023)
556	*Ziziphus mauritiana* Lam. Rhamnaceae	Chitwan, Sinduli, Nuwakot, Sindupalchowk, Makawanpur	Root/fruit juice/paste for fever, indigestion, measles, cough, boils. Pulp fruits edible. (Dangol & Gurung, 2008; Bhattarai & Tamang, 2017; Joshi et al., 2020; Gurung, 2023; Karki et al., 2023)
557	*Ziziphus nummularia* (Burm.f.) Wight & Arn. Rhamnaceae	Chitwan	For ear ache, fever reduction. (Dangol & Gurung, 2008)

Journal of Plant Resources (2025) **23**(1), 156-159
https://doi.org/10.3126/bdpr.v23i1.83360

Short Communication

Wood Anatomy of *Schima wallichii* (DC.) Korth. from Central Nepal

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The genus *Schima* Reinw. ex Blume. is an important genus of the family Theaceae. The genus comprises of altogether than 17 species distributed in different parts of the Tropical & Subtropical Asia (Plants of the World Online [POWO], 2025). In Nepal, the genus *Schima* is represented by single species, *Schima wallichii* (DC.) Korth. (National Herbarium and Plant Laboratories [KATH], 2025; Press et al., 2000; Rajbhandari et al., 2021; Shrestha et al., 2022). The native distribution of Chilaune extends from Central Himalaya to South China and West & Central Malaysia. The species is locally known as Chilaune and form the dominant forest type in subtropical regions in Nepal, predominantly between 1000-2000 m asl. *Schima wallichii* is one of the dominant vegetation components in the subtropical evergreen forest commonly known as the *Schima-Castanopsis* Forest (Chaudhary, 1998; Dobremez, 1976; Forest Research and Training Centre [FRTC], 2021; Suzuki et al., 1991).

Schima wallichii (DC.) Korth. is a large, evergreen, multipurpose, sub-tropical tree (Pearson & Brown, 1932; Suzuki et al., 1991). The bark is gray to dark reddish brown, becoming ruggedly cracked into small, thick angular pieces. It is renowned for its wood which is considered one of the quality timbers in Nepal as well as south Asia. The wood is found to be used to be used in making door and window frames, ploughshares, railway tracts, beams, flooring, and interior, and firewood. It is often cultivated as a shade tree and is used as a pioneer species in the reforestation programs, it benefits through soil and water conservation, has medicinal values, its bark is rich is tannins and is used for dyeing (Tropical Plants Database [TPD], 2025).

Wood is hard, moderately heavy, and medium textured, without characteristic odour and taste; light red to reddish brown; sapwood and heartwood similar, gradual transition from sapwood to heartwood (Gamble, 1972; InsideWood, 2025; Pearson & Brown, 1932; Suzuki et al., 1991; TPD, 2025). But the wood is highly susceptible to insects and fungal attack (Pearson & Brown, 1932). It is easy to work with hand and machine tools and polishes satisfactorily; good-quality plywood can be manufactured from the wood, and it is suitable to produce wood-wool boards (TPD, 2025).

Vigorous increases in global trade and roadways expansion have resulted in over-exploitation of

(a) (b) (c) (d)

Figure 1: Study species: *Schima wallichii*, (**a).** collection of wood samples, (**b).** flowering twig, **(c).** flower close- up, **(d).** fruit

forest resources so the identification and traceability of wood has become very crucial in the present context. Therefore, developing accurate species level identification through its anatomical study seems to be a significant technical prerequisite for the laboratory level identification and treatments of timbers.

Wood samples were collected from Phulchowki hill, Central Nepal during September, 2023 and brought to the KATH Xylarium, boiled at 100ºC in hot air oven for softening and sectioning was done via semi-automatic microtome KDEE 3390. Temporary slides were prepared out of the resulting micro-sections, which were observed under light microscope and fine sections were selected. These sections were then dehydrated in Ethanol series, stained with Safranin and Fast green to prepare the permanent slides. The permanent slides were then studied under Olympus CX43 trinocular compound microscopes at different magnification and photomicrographs were taken by Olympus LC30 camera fitted with 1X adapter.

General properties of wood

Wood is hard, moderately heavy, close grained and without any odour. Heartwood is brown to reddish brown or, greyish brown, without any color stripes. Sap wood is similar to heartwood in terms of color and texture. Growth rings absent or, not very distinct, rarely seen as faint boundaries between earlywood and latewood. Pores evenly distributed, of nearly same size class or, sometimes slightly different in size.

Wood structure and composition

Wood is diffuse porous type. Vessels few, 5-10 vessels per mm^2, exclusively solitary, rarely in radial pairs, evenly distributed. Solitary vessel angular, slightly elliptic to oval in outline; 50-140 µm and 60-110 µm in radial and tangential diameters respectively. Vessel elements 650-1040 µm long; perforation plates scalariform, end walls oblique. Tyloses not seen. But, Pearson and Brown (1932) reported that vessel elements are 430-2990 µm long. However, Chalk (1989) considers the size as well as number of vessels is susceptible to environmental influence.

Vasicentric tracheids usually absent. Fibre-tracheids form the ground mass of the wood, square to polygonal in cross- section. Fibres non- septate, thick walled, pits circular, showing distinct bordered pits. These fibres influence both strength as well as shrinkage of wood (Anoop et al., 2019) due to which the wood of *Schima wallichii* is hard and is difficult to seasoning.

Wood parenchyma diffused, square to polygonal in cross-section, thin walled, axial parenchyma both apotracheal as well as paratracheal; apotracheal parenchyma diffused; paratracheal parenchyma sparse. Individual cells in Axial parenchyma 20-30 µm and 10-25 µm in radial and tangential diameters respectively. Prismatic crystals present in axial parenchyma cells, usually single crystal per cell. Similar features have been described in the InsideWood (2025). Suzuki et al. (1991) reported that prismatic crystals are present in a series of vertically arranged row of square cells. However, Richter & Dallwitz (2000) reported the rare occurrence of two crystals per chamber in axial parenchyma cells.

Rays not visible with the naked eyes, heterogenous; made up of parenchyma; multiseriate as well as uniseriate, multiseriate portions as wide as uniseriate portions of rays. Rays comprise of procumbent cells, upright cells and square marginal cells. Vessel-ray pits with circular or slightly oblique slit-like apertures. Starch deposits are abundant in sapwood (Pearson & Brown, 1932).

Schima wallichii is an important timber tree, that is characterized by the presence hard, durable, diffuse porous wood; exclusively solitary vessels, scalariform perforation plates; non-septate, thick-walled fibres; diffused axial parenchyma, presence of prismatic crystals in chambered axial parenchyma; rays both multiseriate and uniseriate, rays always heterogenous. However, the anatomical features sometimes can be of adaptive value so further studies on the comparative anatomical examination of same plant species existing in different ecological regions would be of great importance.

The authors would like to acknowledge Ms. Jwala Shrestha, Chief, National Herbarium and Plant

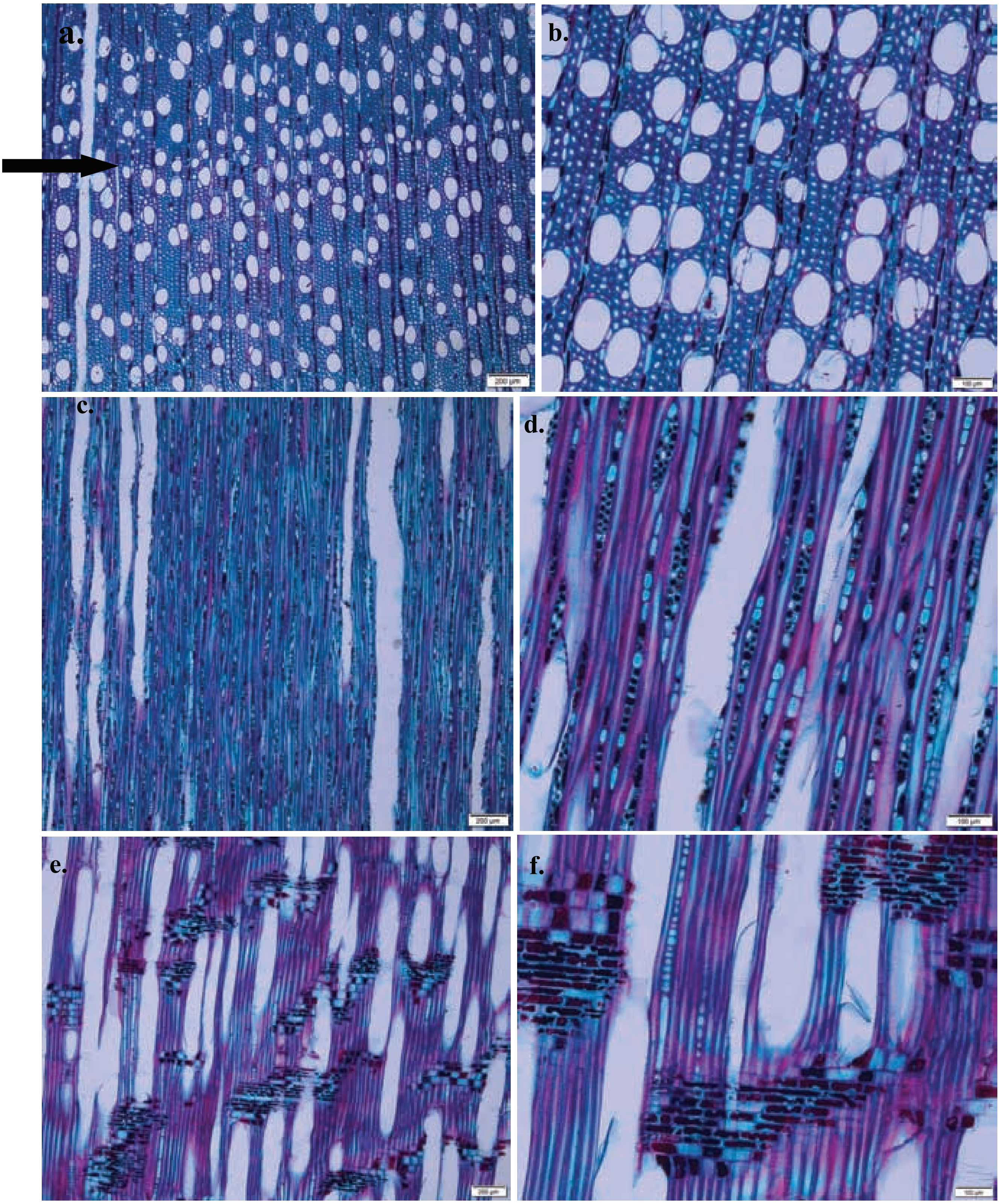

Figure 2: Wood anatomy of *Schima wallichi,* **(a), (b).** Cross-section of wood (TS) showing solitary vessels, vessels angular in cross- section, **(c), (d).** TLS of wood showing both uniseriate and multiseriate rays, **(e), (f).** RLS of wood showing heterogenous rays, Scalariform perforation plates in vessels seen in **(f).** Magnification: a,c,e (4x+1x), b,d,f (10x+1x) magnification. Arrow in figure **(a)** showing the faint growth ring boundary

Laboratories, Godawari, Lalitpur for her direct as well as indirect support and motivation to shape this work in present form. The authors are also thankful to Ms. Srijana Thapa Somai, KATH for her assistance during photomicrography.

References

Anoop, E. V., Rao, R. V. & Mukundan, G. (2019). Wood anatomy and wood property variation in Red Sanders. In Pulliah, T., Balasubramanya, S., & Anuradha, M. (Eds.), *Red Sanders: Silviculture & Conservation* (pp. 131-152). Springer.

Chalk, L. (1989). Vessels. In Metcalfe, C. R., & Chalk, L. (Eds.), *Anatomy of dicotyledons. wood structure and conclusion of the general introduction* (2nd ed.) (Vol. II). Oxford Science Publications.

Chaudhary, R. P. (1998). *Biodiversity in Nepal: Status and conservation*. Craftsman Press.

Dobremez, J. F. (1976). *Le Ne´pal: E´cologie et Bioge´ographie* [Ecology and Biogeography of Nepal]. Centre Nationale de la Recherche Scientifique.

Forest Research and Training Centre. (2021). *Vegetation types of Nepal: A report based on review of literature and expert knowledge. EFTMP Technical Working Document, No.2*. Ecosystem and Forest Types Mapping Program (EFTMP).

Gamble, J. S. (1972). *A manual of Indian timbers*: *An account of the growth, distribution, and uses of the trees and shrubs of India and Ceylon, with descriptions of their wood-structure*. Bishen Singh Mahendra Pal Singh.

InsideWood. (2025). Theaceae *Schima wallichii*. Retrieved on various dates, from https://insidewood.lib.ncsu.edu/search

National Herbarium and Plant Laboratories. (2025). *Schima wallichii* (DC.) Korth. Retrieved on various dates, from http://www.kath.gov.np

Pearson, R. S. & Brown, H. P. (1932). *Commercial timbers of India* (Vol. I) (pp.64-66). A. J. Reprints Agency.

Plants of the World Online. (2025). *Schima* Reinw. ex Blume . Retrieved on various dates, from https://powo.science.kew.org/taxon/urn:lsid:ipni.org:names:39119-1

Press, J. R., Shrestha, K. K. & Sutton, D. A. (2000). *Annotated checklist of the flowering plants of Nepal*. The Natural History Museum.

Rajbhandari, K. R., Rai, S. K. & Chhetri, R. (2021). *A handbook of the flowering plants of Nepal* (Vol. 3). National Herbarium and Plant Laboratories.

Richter, H. G., & Dallwitz, M. J. (2000). Commercial timbers: Descriptions, illustrations, identification, and information. Retrieved on various dates, from https://www-archiv.fdm.uni-hamburg.de/b-online/wood/english/thescwal.htm.

Shrestha, K. K., Bhandari, P., & Bhattarai, S. (2022). *Plants of Nepal* (*Gymnosperms and Angiosperms*). Heritage Publishers & Distributors Pvt. Ltd.

Stainton, J. D. A. (1972). *Forests of Nepal*. John Murray.

Suzuki, M., Noshiro, S., Takahashi, A., Yoda, K., & Joshi, L. (1991). Wood structure of Himalayan plants, II(pp. 5-62). In H. Ohba, & S. B. Malla (Eds.), *The Himalayan Plants* (Vol. 2). University of Tokyo Press.

Tropical Plants Database. (2025). *Schima wallichii* - Useful Tropical Plants. Retrieved on various dates, from https://tropical.theferns.info/viewtropical.php?id=Schima+wallichii

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