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Cover Photo: Hypericum cordifolium and Bistorta milletioides (Dr. Keshab Raj Rajbhandari) Silene helleboriflora (Ganga Datt Bhatt), Potentilla makaluensis (Dr. Hiroshi Ikeda)

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FOREWORD

Nepal is a rich in natural resources with geographic and climatic diversity which culminates into a great diversity of flora and fauna including endemic flora and fauna. Nepal is one of the important places for scientists, naturalists and researchers in unveiling natural treasures. The Department of Plant Resources (DPR) is engaged in developing new methods and technique in plant resources in conservation, sustainable use and equitable sharing of benefits to enhance livelihood of rural people.

Since its establishment in 2016 B.S., this department has been involved in plant exploration, taxonomical identification, ecological studies, mycology, anatomy, ethnobotany, biotechnology, study of active chemical constituents, biological activity of active ingredients, bio-prospecting etc. With our long time dedication and expertise, we have been successful in bringing out the valuable publications such as local flora, fascicles, checklists, MSDS of MAPs and catalogues of flowering as well as non-flowering plants of Nepal. In addition to this department has developed generic guidelines for Good Agriculture Practices (GAP) and Good Cultivation Practices (GCP) of Medicinal and Aromatic Plants (MAPs) as national standard. The department also conducts phytochemical screening researches for bio-prospecting of unexplored plant of Nepal.

Being one of the parties to the Convention on Biological diversity 1992, Nepal Biodiversity Strategy and Action plan 2014-2020 has given emphasis on the publication of all the volumes of Nepal Flora by 2020. Volume 3 and companion volume to the Flora of Nepal have already been published in collaboration with Royal Botanical Garden Edinburgh, UK, Society of Himalayan Botany, Japan, Nepal Academy of Science and Technology (NAST) and Central Department of Botany, Tribhuvan University. Furthermore emphasis is given to the accreditation of laboratories of DPR. The laboratory accreditation is in the process and first assessment of laboratory has been carried out by National Accreditation Body for Testing and Calibration Laboratory (NABL), India.

For dissemination of information on the activities, we conduct seminar, workshops, interactions, publish newsletters and scientific bulletins. I hope the present bulletin of Plant Resource will provide adequate materials to the interested readers, researchers, students and concerned stakeholders.

I would like to thank the painstaking efforts of all the paper contributors, editorial board members, reviewers and staffs of Publicity and Documentation Section of DPR to bring out this publication in this form.

Rajdev Prasad Yadav
Director General
April 2016
Editorial

We are pleased to bring out the present issue of Plant Resource “Bull.Dept.Pl.Res.No. 38”, a continuation of research publication by Department of Plant Resources (DPR). The issue carries a score of peer reviewed articles based on original research. Since this publication intends to highlight on the scope and objectives of our Department, we are aware that the publication should accommodate as many articles as possible so that it represents the work of DPR. And at the same time we are also aware of the need to produce the scientific quality and integrity of the research articles. 20 articles have been incorporated in this issue under different categories like systematic botany, ethnobotanical study, floristic survey, ecology, biotechnology, study of the effect of active chemical constituents of plants on the live animals and biological study (Microbiological). This issue also includes a special article on Endemic Flowering Plants of Nepal. Nepal is rich not only in floristic and faunal diversity but also on endemism due to its phytogeographical diversity.

We encourage our scientists to pursue quality research and contribute to build scientific knowledge on phytochemical screening and pharmaceutical researches for bio-prospecting of unexplored plant of Nepal. The reviewers of the articles published here have contributed much of their knowledge and time to justify the quality of research articles. We heartily thank them all. We duely acknowledge the contribution of contributors for their interest in publishing their valued work in this publication and looking forward to further cooperation and collaboration with other scientific institution. We value the comments and opinion of our contributors and readers. We apologize in advance for any lapses in this issue and at the same time promise to improve the future issues based on your valued input.
Revised checklist to the mycotaxa proposed from Nepal

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Abstract

The proposed mycotaxa accumulated since the works of Berkely (1854) from Nepal are revised based on 156 published papers for their endemism. The papers record about 5 monotypic taxa and 203 species of mycobiota gathered from different regions of Nepalese Himalayan belt. At present, the list shows 131 endemic species. Among them the nomenclature of 120 taxa remains as proposed, while 11 species have undergone synonyms. Sixtyfour species have been recorded to occur in other countries. Two taxa remains invalid.

Key words : Endemic taxa, distribution, synonyms, Nepal Himalaya

Introduction

Nepal Himalaya has been considered as centre for origin, appearance and dominance of number of mycobiota and their distribution pattern between high and low altitude forms, the present number of species is 10 times smaller in comparison to India (Adhikari 1990ab, 2000, 2009, 2014a). Yet the mycobiota Nepal serves as a connecting link with Sino-Japanese, Western and Central Asiatic, North East American, partly North African and mostly Indian subcontinent. The eastern sector shows some of the flora characteristic to South-East Asia, while the rest shows their affinity with the North-West Himalayan elements. Adhikari (2000, 2014ab) also considered 3 mycogeographic regions on the basis of findings of *Amauroderma rugosum* in (Sanghu, Dhankuta) Eastern Himalaya and *Aecidium urticae* in (Bajhang) North-West Himalaya, which supports the view of Stearn (1960) to be divided into 3 phytogeographic regions. The horizontal and vertical distribution of many mycobiota in Nepal coincides well with the phytogeographical division proposed by Dobremez (1972). The alpine zone is rich in many endemic mycobiotypes. Some species from the subtropical and tropical zones include pantropical species. In the dry N.W. Nepal there occur several xerophilous species which are absent from the rest of Nepal; some of them are endemic, others range from Nepal through Middle Asia to W. Europe (Kreisel, 1976).


Till now 721 genera 2467 species 5 gen.nov. and 203 sp. nov. have been reported (Adhikari, 2000, 2009, 2014b) from different regions of Nepalese Himalayan belt. The revised list shows that among 131 endemic species the nomenclature of 120 taxa remains as proposed, while 11 species have undergone nomenclature changes. Sixtyfour species have been recorded to occur in other countries. Two taxa remains invalid.
Materials and methods

This paper is the result of review of 155 papers to find out the prevailing endemic mycobiota of this Himalayan country. Near about 20 species were found unpublished records (not provided here) deposited in different herbaria of the world (for list see Adhikari, 2014b). The year of publication of the proposed new species follows the authors (indicated in the parenthesis). Moreover based on the web data (Mycobank, 2015; Wikipedia, 2013; Species Fungorum, 2015; Catalogue of Life: 2012 Annual Checklist)), different publications [Guzmán & Ramírez-Guillén, (2004), Núñez & Ryvarden (2001), Ramadive (2012), Yang (1997) and Zhuang & Wei (1994, 2002)] and list of species deposited in CAB Herbarium (Meulder, 2006), the species are grouped under monotypic taxa, endemic species, species reported from other countries, nomenclature changes and distribution and invalid taxa (according to ICBN).

Enumeration of species

1. Monotypic taxa
   A. From Nepal
      b. Cladosporothyrium; Cladosporothyrium nepalense Katumoto (1984) - Nepal
   B. From Nepal and other country
      a. Amylaria: Amylaria himalayensis Corner (Balfour-Browne, 1955) – Bhutan (Holotype) and Nepal

2. Endemic species
   1. Aecidium pleurospermae Balfour Browne (1955)
   3. Anixiopsis biplanata Guého & De Vroey (1986)
   15. Cercospora zantedeschiana Singh & Nisha (1973b)
   17. Chroogomphus asiaticus Miller & Aime (2001)
   18. Chrysomyxa taghishae Balfour-Browne (1955)
   21. Coleosporium nepalense Durrieu (1979)
   23. Coniochaeta nepalica Minoura, Morinaga & Muroi (1977)
   25. Dasyscyphus thindii Sharma (1983a)
   27. Dictyostelium aureocephalum Hagiwara (1991)
   29. Dictyostelium gracile Hagiwara (1983)
   31. Dictyostelium medium Hagiwara (1990)
   32. Disciseda ochrochalcea Kreisel (1976)
   33. Eupenicillium nepalense Takada & Udagawa (1983)
   35. Gymnosporangium pamarense Balfour-Browne (1955)
   36. Hamaspora dobremezii Durrieu (1975)
37. Hamaspora viennottii Durrieu (1977b)
41. Lamproderma nigrisplendidum Poelt (1969)
42. Lycogala fuscoviolaceum Onsberg (1973)
43. Lycoperdon altimontanum Kreisel (1976)
44. Lycoperdon elongatum Berk. (1854)
45. Lycoperdon lambinonii var. quercetorum Kreisel (1976)
46. Melampsora ribis Durrieu (1979)
47. Meliola castanopsidis Budathoki & Singh (1994)
49. Meliola santalacearum Budathoki & Singh (1994)
50. Mollisia dhankutae Balfour-Browne (1968)
57. Parrotia melachininensis Balfour Browne (1968)
58. Passalora nepalensis Adhikari & Manandhar (1986)
63. Phellinus politi Ryvarden & Hjortstam (1977)
64. Phellinus subsanfordii Hattori (2002)
66. Phragmidium quinquiloculare var. triseptatum Durrieu (1977)
67. Pleurotus nepalensis Corner (1955 – in Balfour-Browne, 1955; Mycobank, August 2015[update], Index Fungorum lists)
68. Podosporium himalensis Balfour Browne (1968)
70. Pseudocercospora hibiscigena Singh, Singh & Tripathi (1996)
73. Pseudocercospora pileae Singh, Singh & Tripathi (1996)
75. Pseudocercospora urticaearum Verma & Kamal (1989)
76. Pseudocercospora arcuata Singh, Singh, & Bhall (1997)
77. Pseudocercospora woodfordiana Singh, Singh & Bhall (1997)
78. Puccinia annapurnae Durrieu (1979)
80. Puccinia kyangjinensis Ono, Adhikari & Rajbhandari (1988)
81. Puccinia mallae Durrieu (1979)
82. Puccinia manangensis Durrieu (1979)
83. Puccinia mercei Durrieu (1987)
84. Puccinia morduei Adhikari (1998)
85. Puccinia ophiopogonis var. phulchowkiensis Adhikari (1998)
86. Radulum spongiosum Berk. (1854)
87. Ramaria fuscobrunnea Corner (in Balfour-Browne, 1955)
88. Ravenelia microcephala Durrieu (1980)
89. Rhytisma piceum Berk. (1854c)
90. Russula chloroides var. godavariensis Adhikari (1999)
91. **Russula delica** var. **dobremezii** Adhikari (1999)
92. **Russula kathmanduensis** Adhikari (1999)
93. **Russula nepalensis** Adhikari (1990)
94. **Schiffnerula cannabis** McPartland & Hughes (1994)
95. **Secotium himalaicum** Zang & Doi (1995)
96. **Spathularia bifurcata** Otani (1982)
97. **Sporormia nepalensis** Udagawa & Sugiyama (1982)
98. **Stenella mahoniae** Verma, Budhathoki & Kamal (1989)
100. **Stenella rhododendricola** Misra, Srivastava & Kamal (1999)
101. **Stenellopsis nepalensis** Chaudhary & Singh (1996)
102. **Stereum endocrocinum** Berk. (1854b)
103. **Talaromyces convolutus** Udagawa (1993) [Penicillium convolutum Udagawa, anamorph]
104. **Talaromyces emodensis** Udagawa (1993) [Penicillium emodense Udagawa, anamorph]
105. **Talaromyces tardifaciens** Udagawa (1993) [Penicillium tardifaciens Udagawa, anamorph]
106. **Taphrina nepalensis** Otani & Bhandary (1982)
109. **Tretospora himalayana** Chaudhary & Singh (1996)
111. **Uncinula embeliae** Verma, Chand & Kamal (1990)
112. **Uncinula kydiae-calcinae** Verma, Chand & Kamal (1990)
113. **Uromyces dobremezii** Durrieu (1979)
114. **Uromyces langtangensis** Durrieu (1987) [Uremyces amoenus Sydow & Sydow (1906) – in Ono, Adhikari & Kaneko (1995)]. Comment – **Uromyces langtangensis** with teliospores 27-37 x 18-29 μm is larger and very different from **U. amoenus** (India, Nepal, NPParadise Valley, Mount Tacoma, Washington) with teliospores 18-28 x 14-22 μm (Durrieu, 2015, Pers. com.), so retention of **Uromyces langtangensis** Durrieu, is proposed as endemic to Nepal
115. **Uromyces obesus** Durrieu (1987)
117. **Uromyces kathmanduensis** Adhikari (1998)
118. **Uromyces langtangensis** Durrieu (1998)
119. **Xeromphalina aspara** Mass (1992)
120. **Xylaria fistuca** Berk. (1854d)

3. **Species reported from other countries**
   1. **Alternaria longissima** Deighton & Macgarrie (1968) (in Seung-Hun, Mathur and Neergaard, 1982) - Korea
   4. **Anixiopsis stercoraria** (Hansen) Hansen (1897) - Australia and New Guinea (Catalogue of Life: 2012 Annual Checklist)
   7. **Coleosporium pseudocampanulae** Kaneko, Kakishima & Ono (1990, in Zhuang & Wei, 1994) - China
   8. **Coleosporium himalayense** Durrieu (1977a) – India ( in Singh, Khan & Mizra. 1987)
   9. **Dictyostelium clavatum** Hagiwara (1990) - Nepal and Taiwan (Z.Y.Yeh, in BiotaTaiwanica, publication year not given, 2002 web )
   10. **Dictyostelium exiguum** Hagiwara (1977b) - India (in Singh, Khan & Mizra. 1987)
   11. **Dictyostelium exiguum** Hagiwara (1977a) - Nepal and Taiwan (Z.Y.Yeh, in BiotaTaiwanica, publication year not given, 2002 web)
and western North America (Martin & Alexoupulos 1969) and also known from the Antarctic (Ing & Smith 1983) in Stephenson (2003)]


15. Hemitrichia serpula (Scop.) Rost. var. tubiglabra Nann.-Brem. & Yamam. (1990) – China

16. Inonotus hamusetulus Ryv.(1984) (Ranadive, 2013) - India

17. Lentaria macrospora Corner (1968) (in Balfour-Browne, 1968) - Asia

18. Leotia himalayaensis Otani (1982) - China


20. Lycoperdon perlatum var. dobremezianum Kreisel (1976) - India

21. Lycoperdon yetisodale Kreisel (1969) - China


23. Mycovellosiella adinae- cordifoli Kharwar & Narayan (2000) - India


25. Mycovellosiella neri Kharwar & Narayan (2000)- India

26. Peniophora bicornis Ryvarden & Hjortstam (1979) [Nepal (type locality), Gabon, Reunion Island, Singapore, Taiwan in Hjortstam & Ryvarden, 1984; Mycobank, 2015]

27. Phellinus acontextus Ryv. & Hjort. (1984)[in Ryvarden (1984); Hattori (1999); Ranadive (2013)] – India, Japan

28. Phragmidium cinnamomeum Durrieu (1980, in Zhuang & Wei, 1994) - China


32. Puccinia heraclei nepalensis Durrieu (1979)(in Zhuang & Wei, 1994) - China

33. Puccinia nepalensis Barclay & Dietel (1890)(in Zhuang & Wei, 1994) - China

34. Puccinia pilearum Durrieu (1977)( in Zhuang & Wei, 1994) - China


40. Trametes tephroleuca Berk. (1854) - Europe, Japan China, N. America, India and Nepal

41. Veronaea ficina Kharwar & Singh (2004) - India

42. Veronaea grewiicola Kharwar & Singh (2004) - India

43. Veronaea hippocratiae Kharwar & Singh (2004) - India

4. Nomenclature changes and distribution

Taxa proposed (Syn. denoted by = and enclosed in large brackets [ ]) Valid name

A. Endemic to Nepal


103-110, alone has treated *Ophiocordyceps multiaxialis* and *Ophiocordyceps nepalensis* as separate species.


7. [=*Sphaeria nepalensis* Berk. (1854d)] *Valsa nepalensis* (Berk.) Sacc. (1882)


**B. Reported in other countries**


3. [=*Anthraeicoidea nepalensis* Kakishima & Ono (1988)] *Anthraeicoidea disjuncta* (Lira) M. Piatak (2012) – China, India, Nepal

4. [=*Cercospora triloba repentina* Singh & Nisha (1973)] *Cercospora zebrinea* Peck (1877: Hedwigia 16: 124.)-in many countries

5. [=*Dictyostelium magnum* Hagiwara (1983)] *Dictyostelium gigantium* Singh (1947) – India, Nepal


7. [=*Lentinus inquinans* Berk (1854a)] *Lentinus badius* (Berk.)Berk. (1847) – India, Nepal, Malaysia, Thailand, Japan

8. [=*Lentinus nepalensis* Berk. (1854a); *Pocillaria nepalensis* (Berk.) Kuntz.; *Pocillaria velutinus* (Fr.) Kuntz (1891)] *Lentinus velutinus* Fr (1830) – Australia, India, Nepal


12. [=*Polyporus elatinus* Berk.(1854a)]
Revised checklist to the mycotaxa proposed from Nepal

Postia tephroleuca (Fr.) Julich (1982) – Common
13. [= Polyporus flavidus Berk.(1854b)]
Inonotus flavidus (Berk.) Ryv.(1984) - Japan and Taiwan, Nepal
14. [= Polyporus florideus Berk. (1854b)]
Microporus xanthopus (Fr.) Kuntz. (1898) – Common
15. [= Polyporus nepalensis Berk.(1854);
Polyporus menziesii Berk. (1843);
Polyporus corium Berk. (1854);
Polyporus thwaitesii Berk.(1854);
Polystictus nepalensis (Berk.) Cooke (1886); Microporus nepalensis (Berk.) Kuntze (1898) in Wikipedia 2014]
16. [= Polyporus pictilis Berk. ((1854b)]
Tramates versicolor (L.:Fr.)Pilat – Common
17. [= Puccinia commelinae Durrieu (1979);
18. [= Ravenelia pennatae Durr. (1980)]
Ravenelia parasnathii Yadav (1963) – India, Nepal
19. [= Scleroderma nitidum Berkeley (1854b)] Veligaster nitidum (Berk.) Guzman & Tapia (1995)(in Guzmán & Ramirez-Guillén,2004) - Virgin Islands Costa Rica, Cuba, Mexico and Nepal (type locality)]
20. [= Trametes versatilis Berk (Berkeley, 1854)] Trichaptum byssogenum (Jungh.) Ryv. - Europe, N. America and Nepal.
21. [= Ustilago emodensis Berk.(1854c)]
Liroa emodensis (Berk.):Ciferri,(1933) – India, Nepal, China, Japan
22. [= Ustilago endotricha Berk (1854c)]
Farysoprium endotrichum (Berk.) Vánky Mycotaxon. 71:208 (1999) – Australia

5. Invalid taxa

(No diagnostic characters and Latin diagnoses of both genera and species given according to code of ICBN)
1. Epiccospora parasitica Budhathoki gen. et sp. nov. on Osyaris orborea (1994)
2. Fulvioniaceous nepalensis Budhathoki gen. et sp. nov. on Eurya acuminata (1994)

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Revised checklist to the mycotaxa proposed from Nepal

Photographs

Russula kathmanduensis
Russula nepalensis
Russula chloroides var. godavariensis

Russula delica var. dobremezii
Lactarius thakalorum
Amanita cinnamomescens

Suillus adhikarii
Pholiota microspora var. himalensis
Amanita chepangiana

Puccinia adhikarii
Gymnosporangium padmarense
Passalora nepalensis
Eragrostis cilianensis (Poaceae), a new record for Nepal

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Abstract

Eragrostis cilianensis (Poaceae) is reported as a new record for Nepal.

Key words: Eragrostis cilianensis, new record, Nepal.

Introduction

Eragrostis Wolf is a genus belonging to the family Poaceae, tribe Eragrostideae. It is represented by 350 species in the world, distributed in the tropical and subtropical regions and is characterized by annual or perennial herbs with 2- to many-flowered spikelets and 3-veined lemmas (Chen & Peterson, 2006). Fourteen species of Eragrostis have been reported from Nepal (Hara et al., 1978; Press et al., 2000; Rajbhandari, 2010). Recently, a specimen of Eragrostis collected from Dolpa District, West Nepal has been identified as Eragrostis cilianensis. This species has not been reported before from Nepal and is a new addition to the flora of Nepal.

Eragrostis cilianensis (All.) Vign. ex Janch., Mitt. Naturwiss. Vereins Univ. Wien, n. s., 5: 110 (1907). (Fig. 1).

Poa cilianensis All., Fl. Pedem. 2: 246 (1785).

Loosely tufted annual herb; culms 10-35 cm high, erect or geniculate at base, a line of glanss below each node. Leaf sheaths with tubercle hairs; ligules a line of hairs. Leaf blades flat, glabrous, 6–15 × 0.2–0.5 cm. Panicle oblong or pyramidal, 6-12 cm long; branch usually solitary, ascending. Spikelets dark green or gray-green, compressed, oblong or ovate-oblong, 5–7 × 2–2.2 mm, 10–14-flowered. Glumes subequal or lower glume slightly shorter, 1-veined; upper glume 1–3-veined, glandular along middle vein, 2 mm long. Lemmas chartaceous, broadly ovate-oblong, conspicuously 3-veined, lowest lemma 2–2.4 mm long. Palea persistent, oblanceolate, apex rounded, 1–1.5 mm long, ciliolate along keels. Stamens 3; anthers 0.5 mm long. Caryopsis oblong.

Distribution: Tropical and subtropical regions of the world; India, Nepal, Bhutan, China.
Ecology: Occurs on the flat roof of the house.

Fl. & fr.: July-October.


Notes: *Eragrostis ciliacensis* is an annual glandular species with spikelets breaking up from below upwards. *Eragrostis ciliacensis* is closely related to *Eragrostis minor* Host, which has crateriform (bowl-shaped) glands on its pedicels, whereas the pedicels of *Eragrostis ciliacensis* are devoid of such glands.

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We are grateful to Mr. Rajdev Prasad Yadav, Director General and Mr. Sanjeev Kumar Rai, Deputy Director General, Department of Plant Resources, for their encouragement and support.

References


Zephyranthes citrina Baker (Amaryllidaceae), a new record for flora of Nepal

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Abstract

Zephyranthes citrina Baker (Amaryllidaceae) is reported as a new record for flora of Nepal.

Keywords: Zephyranthes citrina, new record, Nepal.

Introduction

Zephyranthes Herbert belonging to family Amaryllidaceae is a genus of about 40 species, are native to tropical, subtropical and warm temperate regions of America (Mabberley, 2002). Two species of Zephyranthes, viz., Zephyranthes candida (Lindl.) Herb. and Zephyranthes carinata Herb. have already been reported from Nepal (Rajbhandari and Baral 2010). While collecting herbarium specimens a specimen of Zephyranthes was collected from Sarlahi district, central Nepal which was later identified at National Herbarium and Plant Laboratories, Godawari, Lalitpur (KATH) as Zephyranthes citrina Baker. This species is a new record for the flora of Nepal, not recorded by Hara et al. (1978), Press et al. (2000), Bista et al. (2001), Rajbhandari and Baral (2010) and Rajbhandari et al. (2015). The three species of Zephyranthes can be distinguished by the following characters.

1a. Stigma divided; perianth pink, with basal tube .........................................................Z. carinata
1b. Stigma capitate; perianth white or yellow, with or without basal tube .......................... 2
2a. Perianth white (sometimes pinkish on the outside), without basal tube........Z. candida
2b. Perianth bright yellow, with basal tube .......... ..........................................................Z. citrina

Zephyranthes eggersiana Urb. in Symb. Antill. 5: 292 (1907)
Common name: Yellow zephyrlily, citron zephyrlily, yellow rain lily.

Perennial herb with bulb. Leaf blade dull green, to 4 mm wide. Spathe 1.6–2.6 cm. Flowers erect; perianth lemon yellow, funnel-shaped, 3.1–5 cm; perianth tube green, 0.7–1 cm, increasing in diameter, less than 1/3 perianth length, ca. 1/2 (1/3–3/4) filament length, less than 1/2 spathe length; tepals rarely reflexed; stamens diverging, in 2 distinctly subequal sets; filaments filiform, subequal, 1.2–2 cm; anthers 5–7 mm; style longer than perianth tube; stigma capitate, usually among or below anthers, not exserted more than 1 mm beyond anthers; pedicel 2.3–4.4 cm, usually longer than spathe. 2n = 48. (Flora of North America Editorial Committee, 2002).

Distribution: Mexico, US, Central and South America, tropical Africa, Malay Peninsula, Southern Asia, China, Japan and India.

Ecology: Natural grassland.

Flowering: July-October.

Specimen examined: Chhatauna, Sarlahi District, 81 m, 2015.10.19, G. Parmar 20151019 (KATH).
Acknowledgements

Author is thankful to Mr. Rajdev Yadav, Director General, Department of Plant Resources and Deputy Director Generals- Ms. Sushma Upadhyaya and Mr. Sanjeev Kumar Rai; and Mr. Ramesh Basnet, Chief, National Herbarium and Plant Laboratories for their encouragement. Author would also like to show appreciation to Dr. Keshab Raj Rajbhandari, Senior taxonomist, for his co-operation and suggestions.

References


Documentation of the Flora of Ramaroshan Wetland Complex, Achham, West Nepal

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Abstract

Present study documents the flowering plants and some cryptogams of the Ramaroshan wetland complex, Achham on exploring the complete rounding the series of 12 lakes. This study compiled 124 species belonging to 55 families. Among them 72 herbs, 24 shrubs, 13 trees and rest of the 3 species are climbers.

Key Words: Wetland flora, Ramaroshan complex, Achham

Introduction

Wetlands are defined as “natural or artificially created areas, such as swamp, marsh, riverine floodplain, lake, water storage area and agricultural land containing water from underground water resources or atmospheric precipitation that may be permanent or temporary, static or flowing and fresh water or saline”. Wetlands are considered as the most productive and dynamic ecosystem of the earth. Nepal is a signatory country of the international wetland convention (Ramsar/Iran) 1971 and ratified it on 17 April 1988. Nepal Biodiversity Strategy 2002 has recognized wetlands as one of the sectoral strategies. Upto now Nepal has 9 wetlands of Ramsar sites. Recently one more from Kaski district (Including Phewa, Begnas, Rupa etc.) is added this year.

Ramaroshan area which lies in the Achham district, the far western region of Nepal. It is 42 km. away from district headquarter of the Achham district, Mangalsain. This area is accessible by gravelled motorable road. It lies in the north –east corner of Achham bordering with Bajura and Kalikot district. It is famous for its 12 wetlands (6 lakes and 6 marshes) and 18 flatlands (www.ramaroshanwetland.com). The wetland complex started from 2350 m to 3792 m above sea level. The popular stream ‘Kailash’ which flows through the middle of the district is originated from this lake area. The conservation of wetlands requires some prerequisites like inventory, responsible organization, legislation and continuous monitoring (Bhandari et al., 1994). The understanding, exploration and documentation of wetland flora from the western part of Nepal are less than the eastern. It may be due to the remoteness and tough geographic features of it (Basnet et al., 2013). So we focused to document the existing plant species.

Fig: Map of study area

Materials and Methods

The present study based on herbarium and information collection during the field visits that were organized twice representing pre-monsoon and post-monsoon in 2015. The authors completely visited these 12 wetland complex by foot along the trail. The aquatic and semi-aquatic plants were
collected up to 5 meter and 100 meter from lakes margin respectively. During field visit, field note, photography and enumeration (Those identified in the field) of the species were done. The collected herbarium specimens were later identified with comparison at the KATH deposited herbarium and consulting standard literatures (Flora of china, Flora of Bhutan, Flora of British India, Flowers of Himalaya). Later the collected specimens were deposited in KATH herbarium after proper identification.

Result and Discussion

Altogether 124 (5 aquatic, 5 semi-aquatic and 102 terrestrial) flowering plants species were recorded belonging to 55 families. Among them, 72 herbs, 24 shrubs, 13 trees, 3 climbers are given in Annex-1. In terms of habit herbs were dominant (64%) followed by shrubs (21%) and trees (11%), finally the least number of species were recorded as climbers (2.67%). The largest families recorded were Asteraceae (11 species), then Poaceae, Rosaceae and Cyperaceae with 7 species each. The families like Polygonaceae and Urticaceae with 5 and 4 species respectively. Lauraceae, Berberidaceae and Caryophyllaceae each family consists of 3 species. Paris polyphylla, Rubia manjith, Iris kemaonensis, Origanum vulgare, Tanacetum dolichophyllum and Taxus contorta are common medicinal plants. The common trees were Aesculus indica, Dodendecana grandiflora, Lindera pulcherrima, Persea odoratissima, Quercus semi carpifolia and Rhododendron arboretum. We record 11 species of pteridophytes. Among them the common are Asplenium ensiforme, Cheilanthes dalhousiae, Drynaria propinqua, Notholaena himalaica, Oleandra wallichii, Goniophlebium argutum, Polystichum piceopaleaceum and Onychium cryptogammoides belonging to the pteridophytes and 1 species of algae were also listed.

The findings of this study partially contribute to the flora of Nepal, green wealth of the country and commitment to the Ramsar Convention, 1971 and the Convention on Biological Diversity, 1992. It also adds value on ecotourism promotion of the wetland and conservation awareness for the local communities.

Conclusion

The study document 112 species of flowering plants 12 species of cryptogams (1 algae and 11 pteridophytes) belonging to 55 families. The highest number of species reported belonging to the family Asteraceae and lowest number of species reported belonging to the families like Violaceae, Valerianaceae, Buxaceae etc. The wetland is dominated by herbs. We hope this study may help to understanding and documenting the floral diversity of mid hills to high altitude wetlands.

Acknowledgement

The authors are grateful to Mr. Ramesh Bahadur Basnet, Chief, National Herbarium and Plant Laboratories, Godawari for his encouragement, opportunity to field visit and continuously inspiring for the preparation of this manuscript. We also acknowledge Mr. Mitra Lal Pathak for supporting the collection of herbarium specimens.

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http://www.ramsar.org 2016/1/24

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www.flraofbritishindia.com

www.floraofbhutan.com

http://www.ramaroshanwetland.com
Fig. 1: Ramaroshan Lakes with vegetation

Fig. 2: *Potamogeton nudosus*

Fig. 3: *Symplocos paniculata*
## Annex

Table 1: Plant species recorded during field visit

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Scientific Name</th>
<th>Family</th>
<th>Habit</th>
<th>Habitat</th>
<th>Collection number</th>
<th>Date of collection</th>
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</table>
Some frequently found cryptogams

<table>
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<th>S.N.</th>
<th>Scientific Name</th>
<th>Family</th>
<th>Habit</th>
<th>Habitat</th>
<th>Collection number</th>
<th>Date of collection</th>
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<td>H</td>
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<td>10.</td>
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<td>EM</td>
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<td>2015.6.6</td>
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<td>11.</td>
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<td>12.</td>
<td>Polystichum piceopaleaceum Tagawa</td>
<td>Dryopteridaceae</td>
<td>H</td>
<td>EM</td>
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</table>

H= Herb, S= Shrub, T= Tree, E= Epiphyte, A= Aquatic, SA= Semi-Aquatic, EM= Emergent, C=Climber, EN=Enumeration
Preliminary Documentation of Basidiomycetous Fungi (Polypores and Mushrooms) found in Bardia National Park and its Buffer Zone Area, Western Nepal

Rajendra Acharya and Gaurav Parmar
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Abstract
The study was conducted for Basidiomycetous fungi especially polypores and mushrooms, found in Bardia national park and its buffer zone area (Shivapur VDC-3 and 7 of upper and lower Bankhet village respectively; Shivapur VDC-5, Bakuwa village and Neulapur VDC-1, Ambreni village) from October 28 to November 3, 2015. Total of 42 species of basidiomycetous fungi were collected. Out of them, three species being at juvenile stage were unidentified and rest of the identified species were from eight orders belonging to 12 families and 23 genera. Polyporales was found to be the dominant order in the study area with 22 species, followed by Thelephorales (five species). Similarly, Polyporaceae was found to be the dominant family represented by 21 species, followed by Thelephoraceae and Xylariaceae, represented by four and three species respectively. Shorea robusta Gaertn., a dominant tree species of the study area, was found to be key host plant for 21 basidiomycetous species, followed by Terminalia alata Roth (six species).

Keywords: Basidiomycetes, fungi, polypores, mushrooms, Bardia national park, buffer zone

Introduction
Basidiomycetes, the Club fungi, is the second largest group of higher fungi. They are mostly saprophytic and some parasitic. The basidiomycetes differ from all other fungi in that they produce the haploid spores called basidiospores at the club shaped basidium during sexual reproduction (Alexopoulos and Mims, 1979). They have highly developed, profusely branched and septate mycelium of two types i.e. primary mycelium and secondary mycelium. The most familiar members of this class are mushrooms, toadstools, rusts, smuts, bracket fungi or polypores, etc. Members of basidiomycetes can secrete cellulose and lignin digesting enzymes. So, they are the best known decomposers of wood.

Polypores, also known as 'bracket' or 'shelf' fungi due to 'shelf-like' fruiting bodies of some species, are tubiferous basidiomycetes. They have minute to large tubes (Miller, 1984). The tubes open to the exterior by means of pores. The spore bearing surface (pore surface) is located on the underside of the pileus. In each species these tube mouths, or pores are of definite size and shape (Overholts, 1953). The reproductive cells (basidia) form a layer on the inner surface of the tubes. The fruiting body (basidiocarp) may be fleshy, leathery, tough, corky or woody. If they are fleshy, they seldom have a central stipe and therefore, do not resemble a gilled mushroom. Typically they lack a stem, and with a few exceptions are hoof shaped (like a horse hoof) to 'resupinate' (lying flat on the substratum on which they are produced. Polypores on the other hand, often have lateral stipes or no stipe at all (Miller, 1984). Their hyphae are mono-, di- or trimatic. They inhabit wood and cause serious decay, and so are generally known as 'wood rotting fungi' (takhetechyau or bahuchhidritchya in nepali) (Adhikari, 1988).

Mushroom can be defined as a macro-fungus with a distinctive fruiting body, which can be either epigeous or hypogeous (Suman and Sharma, 2005). Fungi generally in the class basidiomycetes are commonly called mushrooms, toadstools, gill fungi, or agarics. A mushroom is generally fleshy, spore bearing fruiting body (i.e. basidiocarp) of a fungus, typically produced above ground on soil or on its food source and characterized by heterotrophic mode
of nutrition (Shrestha, 2014). Mushrooms form large 
fruiting bodies visible without the aid of microscope. 
It mostly grows during the rainy season on damp 
rotten logs of woods, trunks of trees, decaying organic 
matter and in a damp soil rich in organic matter.

Nepal is a well famed place for mycodiversity. So 
far 1,150 mushroom species have been reported from 
Nepal (Adhikari, 2012). Among them 157 species 
are reported to be endemic to Nepal (Adhikari, 
2009); 140 species are edible (Adhikari, 2014a), 66 
species are poisonous (Adhikari, 2009) and 75 
species are medicinal (Adhikari, 2009). Christensen 
et al. (2008), though mentioned of 228 edible species 
found in Nepal, but while listing they provided 
the list of 60 species only. The investigation on 
mushrooms of Nepal started since the contribution 
of Lloyd (1808) and Berkeley (1838), ever since 
several papers have been published and several 
botanical expeditions have been done (Aryal and 
Budhathoki, 2013). Among the biotypes of Nepal, 
phanerogamic floral diversity has been studied 
immensely but the study on cryptogamic flora, 
especially mycodiversity has got less attention 
(Adhikari, 2012). Mushrooms generally prefer wet 
region over dry region for its habitat resulting in its 
high diversity in the central and eastern Nepal as 
compared to the western Nepal.

It has been observed that intense mycological 
exploration and investigations has been done in 
central Nepal as compared to eastern and western 
Nepal (Adhikari, 2000). Moreover, work on 
ymycological exploration and investigation from low 
land Terai region is less as compared to mountain 
and hilly region. Therefore, present study was 
undertaken to document the uninvestigated 
ymycodiversity of Bardia national park and its buffer 
zone area which is situated in the less explored region 
of western Nepal.

Materials and Method

Study area

Bardia national park is situated in Bardia district of 
western Nepal. It covers an area of 968 sq. km. and 
its surrounding area covering 328 sq. km is 
designated as a buffer zone. The buffer zone includes 
altogether 20 village development committees 
(VDCs) around the park. Forest area occupies 59% 
of the buffer zone; and total cultivated area in the 
buffer zone is 41% (Paudel, 2015). Altitude varies 
from 152 m at Manau Ghat to 1,441 m at Sukarmala, 
the highest point of the Churiya range. Mean annual 
rainfall at Chisapani at the foot of the Churiya is 
2,230 mm, and at Gularia to the south of the park is 
1,560 mm. Climate of the study area is typically 
tropical with hot climate. Temperature during the 
hot season rises upto a maximum of 45°C and falls 
down to 5°C during the winter season (Paudel, 2015).

About 70% of the park is covered with dominantly 
sal forest with a balanced mixture of grasslands, 
savannah and riverine forest (Chaudhary, 1998). 
Altogether 839 species of flora were estimated in 
the park (BPP, 1995). One hundred and seventy three 
species of vascular plants are recorded in the park: 
six pteridophytes, one gymnosperm, 140 dicots and 
26 monocots (Bhuju et al., 2006). Upadhyay (2005) 
classified seven major vegetation types in the park. 
These are: sal forest (Shorea robusta, Terminalia 
ala and Buchanania latifolia), khair-sissoo forest 
(Dalberiga sissoo and Acacia catechu), moist 
riverine forest (Syzygium cumini, Mallotus 
philippensis, Bombax ceiba), mixed hardwood 
forest (Adina cordifolia, Casearia tomentosa, 
Mitragyna parviflora), wooded grassland forest 
(Saccharum spontaneum, Imperata cylindrica, 
Erithrina ravennae), Phantas (open grassland) and 
Flood plain grassland (Saccharum spontaneum, S. 
benghalensis, Phragmatis karka and Arundo donax).
Collection and Identification

The park and its buffer zone area (Shivapur VDC-3 and 7 of upper and lower Bankhet village respectively; Shivapur VDC-5, Bakuwa village and Neulapur VDC-1, Ambreni village) were extensively explored for basidiomycetous fungi from October 28 to November 3, 2015. Altogether 42 species of basidiomycetous fungi were collected from nature in the study area. The species collected were well air dried in the shade and packed in paper envelops with proper tag numbers. The species found in the soil were collected carefully by digging with the help of a digger. Other specimens which were found to grow on fallen or rotten branches/wooden logs, branches or trunks of dying or dead plants; or trunks of living plants were collected along with their host plant by cutting with the help of saw. During collection, at least one basidiocarp was left for their spore dispersal.

Photographs of all the species were taken in their natural habitat prior to collection. The habitat/substrate including ecological parameters viz. altitude, vegetation composition, soil type was recorded. The paper envelops were brought to National Herbarium and Plant Laboratories (KATH), Godawari for identification and making herbarium specimens. The identifications were done following key identifying characters of relevant literatures (Alexopoulos and Mims, 1979; Dickson and Lucas, 1979; Pacioni, 1981; Svérèek, 1983; Miller, 1984; Adhikari, 2014). It was also identified by tallying photographs of the relevant literatures and cross checking the collected specimens to that of identified herbarium specimens deposited at Mycology section of National Herbarium and Plant Laboratories. Some species were also identified seeking the help of expert of Mycology. The nomenclature of all the identified species follows Adhikari (2012, 2014).

Results and Discussion

Total of 42 species of basidiomycetous fungi from eight orders belonging to 12 families and 23 genera were collected from the study area (Table 1). During collection, at least one basidiocarp was left for their spore dispersal which support sustainable and scientific collection practice (Adhikari, 2000). Although distribution of macro-fungal species is low in hot and dry season, collection of basidiomycetous fungi was carried out during autumn season for the species commonly found in this season rather than rainy season resulting in fewer collections. Most of the collected basidiomycetous fungi are the members of polyporales. Polyporales, the dominant order, in the study area with 22 species was followed by Thelephorales (five species). Similarly, Polyporaceae was found to be the dominant family represented by 21 species. It was followed by Thelephoraceae and Xylariaceae, represented by four and three species respectively.

Table 1: List of Basidiomycetous fungi collected from Bardia national park and its buffer zone area

<table>
<thead>
<tr>
<th>S. N.</th>
<th>Name of Species/Collection No.</th>
<th>Family</th>
<th>Order</th>
<th>Altitude</th>
<th>Host Plants/Substrates</th>
<th>Locality</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Auricularia auricular-judea</em> (Bull.) Quel. 834B</td>
<td>Auriculariaceae</td>
<td>Auriculariales</td>
<td>158 m</td>
<td>Log of locally called 'Guthil' (<em>Trewia nudiflora</em> L.)</td>
<td>Shivapur-3, upper Bankhet</td>
</tr>
<tr>
<td>2</td>
<td><em>Coriolus hirsutus</em> (Fr.) Quel. 808B</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>160 m</td>
<td>Pole of <em>Shorea robusta</em> Gaertn.</td>
<td>Bardia national park near Shivapur-3, upper Bankhet</td>
</tr>
<tr>
<td>3</td>
<td><em>Dacrymyces palmatus</em> (Schw.) Bres. 842B</td>
<td>Dacrymycetaceae</td>
<td>Dacrymycetales</td>
<td>160 m</td>
<td>Log of <em>Dalbergia sissoo</em> Roxb. ex DC.</td>
<td>Shivapur-3, upper Bankhet</td>
</tr>
<tr>
<td>4</td>
<td><em>Daedielopsis confagosa</em> (Bolt.: Fr.) Schr. var. confagosa 807B</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>160 m</td>
<td>Log of <em>Terminalia alata</em> Roth</td>
<td>Shivapur-3, upper Bankhet</td>
</tr>
<tr>
<td>5</td>
<td><em>Daldinia concentrica</em> (Bolt.: Fr.) Cos. &amp; de Not. 814B</td>
<td>Xylariaceae</td>
<td>Xylariales</td>
<td>160 m</td>
<td>Branch of living tree of <em>Morus australis</em> Poir.</td>
<td>Shivapur-7, lower Bankhet</td>
</tr>
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<td>ID</td>
<td>Species Name</td>
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<td>Order</td>
<td>Habitat Details</td>
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<td>Favolus alveolaris (D.C. ex Fr.) Quel.</td>
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<td>Polyporales</td>
<td>Electric pole of Shorea robusta Gaertn.</td>
<td>Shivapur-7, lower Bankhet</td>
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<td>8</td>
<td>Ganoderma applanatum (Pers.) Pat.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Shorea robusta Gaertn.</td>
<td>Shivapur-7, lower Bankhet</td>
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<td>9</td>
<td>Heterobasidion annosum (Fr.) Bref.</td>
<td>Bonderzewiaceae</td>
<td></td>
<td>Trunk of living tree of Shorea robusta Gaertn.</td>
<td>Shivapur-3, upper Bankhet</td>
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<tr>
<td>10</td>
<td>Hexagonia sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Mangifera indica L.</td>
<td>Bardia national park near Shivapur-7, lower Bankhet</td>
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<td>11</td>
<td>Hexagonia sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Shorea robusta Gaertn.</td>
<td>Neulapur-1, Ambreni</td>
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<td>12</td>
<td>Innonotus hispidus (Fr.) Karst.</td>
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<td>Polyporales</td>
<td>Pole of Shorea robusta Gaertn.</td>
<td>Shivaipur-5, Bakuwa</td>
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<tr>
<td>13</td>
<td>Lenzites betulina (L.) Fr.</td>
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<td>Neulapur-1, Ambreni</td>
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<td>14</td>
<td>Microporus xanthopus (Fr.) Kuntze</td>
<td>Hymenochaeteae</td>
<td>Hymenochaetales</td>
<td>Dead bark of log of Shorea robusta Gaertn.</td>
<td>Shivaipur-3, upper Bankhet</td>
<td></td>
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<tr>
<td>15</td>
<td>Microporus verninctpes (Perk.) Kuntze</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
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<td>Neulapur-1, Ambreni</td>
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<td>16</td>
<td>Oxyoporus populimus (Schw.: Fr.) Donk.</td>
<td>Schizophoraceae</td>
<td>Hymenochaetales</td>
<td>Dead branch of living tree of Terminalia alata Roth</td>
<td>Neulapur-1, Ambreni</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>Phellinus sp.</td>
<td>Hymenochaeteae</td>
<td>Hymenochaetales</td>
<td>Trunk of dying tree of Terminalia alata Roth</td>
<td>Neulapur-1, Ambreni</td>
<td></td>
</tr>
<tr>
<td>18</td>
<td>Phelloden niger (Fr.) Karst.</td>
<td>Bankeraceae</td>
<td>Thelephorales</td>
<td>Log of Dalbergia sissoo Roxb. ex DC.</td>
<td>Shivaipur-3, upper Bankhet</td>
<td></td>
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<tr>
<td>19</td>
<td>Polyporus sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Terminalia alata Roth</td>
<td>Shivaipur-3, upper Bankhet</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>Polyporus sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Grow on dead branch of living tree of Holarrhena pubescens Wall. ex G. Don</td>
<td>Shivaipur-7, lower Bankhet</td>
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<tr>
<td>21</td>
<td>Polyporus sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Dalbergia sissoo Roxb. ex DC.</td>
<td>Shivaipur-3, upper Bankhet</td>
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<tr>
<td>22</td>
<td>Polyporus sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Shorea robusta Gaertn.</td>
<td>Shivaipur-3, upper Bankhet</td>
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</tr>
<tr>
<td>23</td>
<td>Polyporus sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Dead bark of log of Shorea robusta Gaertn.</td>
<td>Shivaipur-3, upper Bankhet</td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>Polyporus squamosus Michel. Fr.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Terminalia alata Roth</td>
<td>Shivaipur-3, upper Bankhet</td>
<td></td>
</tr>
<tr>
<td>25</td>
<td>Polyporus versicolor (L.) Fr.</td>
<td>Polyporaceae</td>
<td></td>
<td>Stump of Terminalia alata Roth</td>
<td>Bardia national park near Shivapur-3, upper Bankhet</td>
<td></td>
</tr>
<tr>
<td>26</td>
<td>Pycnoporus cinnabarinus (Jacq.: Fr.) Karst.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Psidium guajava L.</td>
<td>Neulapur-1, Ambreni</td>
<td></td>
</tr>
<tr>
<td>27</td>
<td>Pycnoporus coccineus (Fr.) Bond. &amp; Singer</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>Log of Shorea robusta Gaertn.</td>
<td>Neulapur-1, Ambreni</td>
<td></td>
</tr>
<tr>
<td>No.</td>
<td>Species Name</td>
<td>Family</td>
<td>Order</td>
<td>Elevation (m)</td>
<td>Location Description</td>
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<td>-----</td>
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<td>------------------</td>
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<td>-------------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>28</td>
<td><em>Pycnoporus</em> sp.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>160</td>
<td>Log of locally called 'Guthil' (<em>Trevis nudiflora</em> L.) Shivapur-7, lower Bankhet</td>
<td></td>
</tr>
<tr>
<td>29</td>
<td><em>Schizophyllum commune</em> (Fr.) Fr.</td>
<td>Schizophyllaceae</td>
<td>Agaricales</td>
<td>168</td>
<td>Log of <em>Terminalia alata</em> Roth Neulapur-1, Ambreni</td>
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<tr>
<td>30</td>
<td><em>Spongipellius delectans</em> (Peck) Murr.</td>
<td>Polyporaceae</td>
<td>Polyporales</td>
<td>160</td>
<td>Fallen branch of <em>Populus ciliate</em> Wall. ex Royle Shivapur-3, upper Bankhet</td>
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<td>31</td>
<td><em>Stereum rugosum</em> (Pers.: Fr.) Fr.</td>
<td>Stereaceae</td>
<td>Russulae</td>
<td>159</td>
<td>Dead bark of log of <em>Shorea robusta</em> Gaertn. Bardi national park, near Shivapur-7, lower Bankhet</td>
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<tr>
<td>32</td>
<td><em>Stereum</em> sp.</td>
<td>Stereaceae</td>
<td>Russulae</td>
<td>157</td>
<td>Log of <em>Shorea robusta</em> Gaertn. Shivapur-7, lower Bankhet</td>
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<td>33</td>
<td><em>Trametes hirsuta</em> (Fr.) Pilat.</td>
<td>Thelephoraceae</td>
<td>Thelephorales</td>
<td>160</td>
<td>Log of <em>Acacia catechu</em> (L. f.) Willd. Bardi national park near Shivapur-3, upper Bankhet</td>
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<td><em>Trametes sp.</em></td>
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<td>Thelephorales</td>
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<td><em>Trametes pubescens</em> (Schum.: Fr.) Pilat.</td>
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<td>38</td>
<td><em>Xylaria furcata</em> Fr.</td>
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<td>Xylariales</td>
<td>161</td>
<td>Log of <em>Shorea robusta</em> Gaertn. Shivapur-7, lower Bankhet</td>
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<td>39</td>
<td><em>Xylaria nigripes</em> (K.I.) Sacc.</td>
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<td>Xylariales</td>
<td>130</td>
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<td>Substrate unknown Shivapur-3, upper Bankhet</td>
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<td>Unidentified2</td>
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<td></td>
<td>162</td>
<td>Log of locally called 'Guthil' (<em>Trevis nudiflora</em> L.) Shivapur-3, upper Bankhet (Khokharapur tole)</td>
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<td>42</td>
<td>Unidentified3</td>
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<td>158</td>
<td>Log of <em>Shorea robusta</em> Gaertn. Shivapur-7, lower Bankhet</td>
<td></td>
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</tbody>
</table>

**Figure 2:** Orders representing number of species in the study area

**Figure 3:** Families representing number of species in the study area
Polypores were the most common and were found to grow on dead woods, fallen logs, stumps, rotten branches but some such as *Daldinia concentrica* (Figure 5), *Fomes fomentarius* (Figure 6), *Polyporus* sp. were found to be very common in the study area. *Shorea robusta* Gaertn. which is the dominant tree species of the study area was found to be key host plant for 21 basidiomycetous species. It was followed by *Terminalia alata* Roth which was found to host six species.

Figure 5: *Daldinia concentrica* (Bolt.: Fr.) Cos. & de Not.

Figure 6: *Fomes fomentarius* (L.: Fr.) Kicks.

*Heterobasidion annosum*, etc. were found on dead branches of trees and trunks of living trees. Out of 42 fungal species, *Daediolopsis confragosa* var. *confragosa* (Figure 7), *Pycnoporus cinnabarinus* (Figure 8), *Xylaria fuscata* (Figure 9), *Trichaptum byssogennum* (Figure 10), *Trametes* sp. and *Xylaria furcata* Fr.
All the host plants or substrates of the fungal species were identified except one since it was almost rotten old wooden log. On other hand, three fungal species, out of 42, could not be identified as the collected species were at juvenile stage.

Conclusion

Total of 42 species of basidiomycetous fungi were collected. Out of them, three species being at juvenile stage were unidentified and rest of the identified species were from eight orders belonging to 12 families and 23 genera were collected from selected study area of Bardia national park and its buffer zone area. Polyporales and Polyporaceae were the dominant order and family respectively. *Shorea robusta* Gaertn. was found to be the major host plant for 21 basidiomycetous species.

Acknowledgements

Authors would like to express gratitude to Mr. Ramesh Basnet, Chief, National Herbarium and Plant Laboratories, Godawari, Lalitpur for encouragement and Dr. Mahesh Adhikari, senior mycologist, for suggestions and cooperation in the identification of fungal species. Authors would also like to appreciate Ms. Basanti Kumpakha, GIS officer, National Trust for Nature Conservation, Khumaltar, Lalitpur for designing map of the study area.

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Documentation of Plant diversity Conserved in Botanical Gardens of Makwanpur, Nepal

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Abstract
This study documents the plants conserved in three Botanical Gardens (BGs) of Makwanpur district. All record plants are in living condition in field and herbarium also prepared and deposited in KATH herbarium. Brindavan BG, Mountain BG and Tistung BG comprise 302 species, 190 species and 138 species respectively. Total 510 plant species were recorded from 391 genera and 130 families. Among them 409 medicinal, 74 ornamental, 35 threatened and 12 protected plant species. BGs are playing important role in conservation of genetic resources of local as well national flora of Nepal.

Keywords: Botanical Garden, Makwanpur, Brindavan, Daman, Tistung, ex-situ, in-situ conservation

Introduction

Among the 11 Botanical Gardens (BGs) of Nepal, three Botanical Gardens (Brindavan BG, Mountain BG and Tistung BG) are in Makwanpur district. This district lies between 84º41' to 84º35' E longitude and 27º21' to 27º24' N latitude, cover total area of 2390 sq km. Its altitude ranges from 166m (at Hathidhunga, Raigaun) to 2584m (at Simbhanjyang, Daman), (Singh, 2003). Due to wide variation in altitude, it harbors plants of tropical, sub-tropical, temperate and sub-alpine vegetation zones (Stainton, 1972). So, three BGs were established for the ex-situ and in-situ conservation of pteridophytes, gymnosperms, orchids, succulents, cactus, medicinal, ornamental, rare, endangered, threatened and protected plants. The main purpose of these BGs is to carry scientific research, collection of herbarium, identification of plants, technology development and educational awareness as well as conservation of genetic resources (Singh, 2003, Lamichhane et al, 2014). Brindavan BG was established in 2019 BS as a medicinal herbal garden under the Department of Plant Resources formerly named as Department of Medicinal Plants. It is located at an altitude of 350-450 m in tropical zone of Churiya range of Central Nepal, Hetauda. It is surrounded by Sal (Shorea robusta Gaertn.) forest and deciduous riverine forest. Similarly, Mountain BG established in 2022 BS, is located at an altitude of 2320 m in Daman area, and is surrounded by Pine, Oak and Rhododendron forest. It harbors generally plants of high mountains with Temperate and Subalpine climate. Likely, Tistung BG was established in 2022 BS, and is located at an altitude of 1900m in Tistung area, and is surrounded by Pine forest (DPRO, 2014, Chapagain et al, 2015)

Materials and Methods

Plants were recorded from three BGs, which were collected from different parts of Makwanpur district and some other places of Nepal (DPRO, 2014, Chapagain et al, 2015). Some ornamental species were exotic too. All plants are in living condition in field and herbarium also prepared and deposited in KATH Herbarium, Godawari. Plants were identified and their uses recorded with help of related literatures (Stainton, 1988, Polunin and Stainton, 1987, Shrestha and Joshi, 1996, Manandhar, 2002, Baral and Kurmi, 2006, Bhattarai and Ghimire, 2006, DPR, 2007, Raskoti, 2009).

Result and Discussion

Diversity

The study recorded 510 plant species which belongs to 391 genera and 130 families (Appendix-1). Among them 218 (42.7%) are herb, 114 (22.4%) are shrub, 152 (29.8%) are tree and 26 (5.1%) are...
climber in habit. Out of them 409 species are medicinal (170 herb, 87 shrub, 132 tree and 20 climber) (Figure-1), 74 ornamental species, 10 invasive species and 27 other species. Among the three BGs, Brindavan BG, harbors 302 species, Mountain BG, harbors 190 species and Tistung BG, harbors 138 species of plants (Appendix 1). Among the below listed plant various parts are used for different medicinal and other purposes viz. 110 sp. for leaves, 96 sp. roots, 66 sp. flowers, 55 sp. fruits, 25 sp. stems, 83 sp. bark, 11 sp. rhizome, 21 sp. seeds, 70 sp. whole plants and some other parts (Appendix-1).

Among the 130 families, Asteraceae is the largest family with 38 species, which followed by Fabaceae

Table 1: Medicinal plants prioritized for research and development

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Scientific name</th>
<th>Nepali name</th>
<th>Family</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aconitum spicatum (Bruhl) Stapf</td>
<td>विष</td>
<td>Ranunculaceae</td>
</tr>
<tr>
<td>2</td>
<td>Acorus calamus L.</td>
<td>बोधी</td>
<td>Araceae</td>
</tr>
<tr>
<td>3</td>
<td>Asparagus racemosus Wild. *</td>
<td>करीसी</td>
<td>Asparagaceae</td>
</tr>
<tr>
<td>4</td>
<td>Azadirachta indica A. Juss.</td>
<td>नीम</td>
<td>Meliaceae</td>
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<tr>
<td>5</td>
<td>Bergenia ciliata (Haw.) Sternb.</td>
<td>पापाबंड</td>
<td>Saxifragaceae</td>
</tr>
<tr>
<td>6</td>
<td>Cinnamomum glaucescens (Nees.) Nand.Mazz*</td>
<td>सुगन्ध कीकिला</td>
<td>Lauraceae</td>
</tr>
<tr>
<td>7</td>
<td>Cinnamomum tamala (Buch.-Ham.) Nees &amp; Eberm</td>
<td>तेजपत</td>
<td>Lauraceae</td>
</tr>
<tr>
<td>8</td>
<td>Dioscorea deltoidea Wall. ex Griseb.</td>
<td>बन तरुल</td>
<td>Dioscoreaceae</td>
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<td>9</td>
<td>Gaultheria fragrantissima Wall.</td>
<td>दोळपाण</td>
<td>Ericaceae</td>
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<tr>
<td>10</td>
<td>Juglans regia L.</td>
<td>ओखर</td>
<td>Juglandaceae</td>
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<tr>
<td>11</td>
<td>Lichens</td>
<td>भागु</td>
<td>Helvellaceae</td>
</tr>
<tr>
<td>12</td>
<td>Morchella conica Pers.</td>
<td>दुध पारा</td>
<td>Euphorbiaceae</td>
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<td>13</td>
<td>Phyllanthus emblica L.</td>
<td>अमला</td>
<td>Piperaceae</td>
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<td>Piper longum L.*</td>
<td>पिपला</td>
<td>Apocynaceae</td>
</tr>
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<td>15</td>
<td>Rauvolfia serpentina(L.) Benth ex Kurz*</td>
<td>सर्पागन्धा</td>
<td>Polygonaceae</td>
</tr>
<tr>
<td>16</td>
<td>Rheum australe D. Don</td>
<td>पदम चाल</td>
<td>Gentianaceae</td>
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<td>Rubia manjith Roxb. ex Fleming</td>
<td>मंजिठ</td>
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<td>18</td>
<td>Sapindus mukorossi Gaertn.</td>
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<td>Swertia chirayita (Roxb. ex Fleming) Karsten*</td>
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<td>Valeriana jatamansi Jones*</td>
<td>सुगन्ध चवाण</td>
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<td>24</td>
<td>Zanthoxylum armatum DC.*</td>
<td>तिमुर</td>
<td>Rutaceae</td>
</tr>
</tbody>
</table>

(*-priorized for agro-technology development)
Prioritized medicinal plants

DPR (2011) has prioritized 30 medicinal plants for research and development in Nepal. These 3 BGs of Makwanpur comprises 24 species among them (Table-1). Similarly, out of 12 species prioritized for agro-technology development, 9 species are conserved and promoted for research in these BGs (Table-1).

Conclusion

Since its establishment, Brindavan BG has been used to conserve genetic resources of more than 302 plant species. Similarly, Mountain BG, Daman and Tistung BG have conserved 190 and 138 species respectively. Among the total 510 species conserved, 409 species medicinal, 74 species ornamental, 35 species threatened, 12 protected species were conserved and promoted for agro-medicinal farming. BGs have been practiced for the in-situ and ex-situ conservation techniques for conservation of rare, endangered, economically important medicinal and ornamental plants. It has conducted training for agro-farming of medicinal plants and researches on aromatic plants.

Aknowledgements

We are thankful to Mr. Rajdev Pd. Yadav, Director General, Mrs. Sushma Upadhyaya, Deputy Director General, Mr. Sanjeev Kumar Rai, Deputy Director General, Department of Plant Resources and Dr. Akhileshwor Lal Karna, Regional Director, Central Regional Forest Directorate, Hetauda for their encouragement. Our sincere thanks to all staff of District Plant Resources Office, Makwanpur for their kind help.

References


## Appendix 1: Plants of Brindavan, Mountain and Tistung Botanical Garden of Makwanpur

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Scientific name</th>
<th>Nepali name</th>
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<th>Lf</th>
<th>Use</th>
<th>Part</th>
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<td>St</td>
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<td>Med</td>
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<td>Fabaceae</td>
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<td>Med</td>
<td>Fr</td>
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<td>Red cattail</td>
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<td>Orn</td>
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<td>Acer pectinatum Wall. ex Pax.</td>
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<td>Lf</td>
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<td>Med</td>
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<td>BT</td>
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<td>Br</td>
<td>B</td>
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<td>B</td>
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<td>Med</td>
<td>Rb</td>
<td>B</td>
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<td>Ardisia solanacea Roxb.</td>
<td>Damai fal</td>
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<td>Wb</td>
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<td>Ardisia tinctoria Roxb.</td>
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<td>Fr</td>
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<td>Argemone mexicana L.</td>
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<td>Rf, Lf</td>
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<td>Tuber</td>
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<td>Artemisia dubia Wall. ex Besser</td>
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<td>Kagapi ghans</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>467</td>
<td>Terminalia alata Heyne ex Roth</td>
<td>Salji</td>
<td>Combretaceae</td>
<td>T</td>
<td>M, ed</td>
<td>Br, Lf</td>
<td></td>
<td></td>
</tr>
<tr>
<td>468</td>
<td>Terminalia bellirica (Garten.) Roxb. Barro</td>
<td>Combretaceae</td>
<td>T</td>
<td>M, ed</td>
<td>Br, Lf</td>
<td></td>
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</tr>
<tr>
<td>469</td>
<td>Terminalia chebula Retz.</td>
<td>Harro</td>
<td>Combretaceae</td>
<td>T</td>
<td>M, ed</td>
<td>Br, Lf</td>
<td></td>
<td></td>
</tr>
<tr>
<td>470</td>
<td>Terminalia catappa L.</td>
<td>Kajagaj badam</td>
<td>Combretaceae</td>
<td>T</td>
<td>M, ed</td>
<td>Br, Lf</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Rajesh Tamang and N.H. Chapagain
### Appendix 2: Plants of threaten category found in BGs of makwanpur

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Scientific name</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Abies spectabilis (D. Don) Mib.</td>
<td>NT, Protected</td>
</tr>
<tr>
<td>2</td>
<td>Acacia catechu (L.) Willd.</td>
<td>CT, Protected</td>
</tr>
<tr>
<td>3</td>
<td>Aconitum spicatum (Haw.) Sternb.</td>
<td>CT</td>
</tr>
<tr>
<td>4</td>
<td>Alstonia scholaris</td>
<td>NT, Protected</td>
</tr>
<tr>
<td>5</td>
<td>Asparagus racemosus</td>
<td>CT</td>
</tr>
<tr>
<td>6</td>
<td>Bergenia ciliata (Haw.) Stemmb.</td>
<td>CT</td>
</tr>
<tr>
<td>7</td>
<td>Bombax ceiba L.</td>
<td>Protected</td>
</tr>
<tr>
<td>8</td>
<td>Butea monosperma (Lam.) Kuntze</td>
<td>EN</td>
</tr>
<tr>
<td>9</td>
<td>Choerospondias axillaris (Roxb.) B. L. Burtt &amp; A. W. Hill</td>
<td>Rare</td>
</tr>
<tr>
<td>10</td>
<td>Cinnamomum glauescens (Nees.) NandMazz</td>
<td>Protected</td>
</tr>
<tr>
<td>11</td>
<td>Crataeva unicollarius Buch.-Ham.</td>
<td>Rare</td>
</tr>
<tr>
<td>12</td>
<td>Curculigo orchioides Gaertn.</td>
<td>Vul (CAMP)</td>
</tr>
<tr>
<td>13</td>
<td>Cycas pectinata Griff.</td>
<td>CITES II, EN</td>
</tr>
<tr>
<td>14</td>
<td>Dalbergia latifolia Roxb.</td>
<td>Vul, Protected</td>
</tr>
<tr>
<td>15</td>
<td>Dioscorea deltoidea Wall. ex Griseb.</td>
<td>CITES II, CT</td>
</tr>
<tr>
<td>16</td>
<td>Elaeocarpus sphaericus (Gaertn.) K. Schum.</td>
<td>Vul</td>
</tr>
<tr>
<td>17</td>
<td>Juglans regia L.</td>
<td>Protected</td>
</tr>
</tbody>
</table>

(Acronyms: NT=Near threatened, CT=Commercially threatened, Vul=Vulnerable, EN=Endangered)


**Puccinia thaliae** Dietel (Uredinales) parasitic on **Canna indica** L.: a new record from Nepal

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and
UMR Evolution et Biodiversité, Université Paul Sabatier, Toulouse, France

**Abstract**

*Puccinia thaliae* Dietel, the rust, found parasitic on cultivated *Canna indica* L., was collected in Bhanimadal, Rudreswar tole, Lalitpur, which is new record from Nepal.

**Key words** – Canna, Rust, Nepal

**Introduction**

The horticultural plant *Canna* L. parasitized by *Puccinia thaliae* Dietel (the fungal disease rust: Urediniomycetes, Uredinales, Pucciniaceae) produces pustules on the both surface of plant’s leaves. The rust attacks severely causing yellow to yellow brown spots on the upper leaf-surface, which coalesce and turn to brown-to-black as the disease increases. It infects the stems also.

*Canna indica* L. (Sarbada in Nepali) is a popular ornamental plant cultivated everywhere in Nepal. It is native to tropical South America. Kaur, Rush, Ferrin & Aime (2011) confirmed the pathogen parasitic on this plant as *Puccinia thaliae* Dietel by rDNA sequencing. *Puccinia thaliae* has been reported previously from Hawaii, India and South Africa (Gardner & Martinez, 1985; Gardner & Hodges, 1989; Jeeva et al., 2004; van Jaarsveld et al., 2006; Nelson, 2013; Neo & Tham, 2009).


The present author accidently found this rust at Bhanimandal, Lalitpur, parasitizing the *Canna* plant. The rust was studied under the microscopic magnification of 10 x 40. The specimen is deposited in National Herbarium and Plant Laboratories, Kathmandu (KATH), Nepal.

**Description**


Aecia not found. Uredinia pustules 1-2 mm in diameter, golden yellow to yellowish brown, subepidermal, erumpent, irregular-shaped, uredinia on both leaf surfaces, sori scattered to covering the entire leaf with coalescing pustules. Urediniospores 26 – 40 x 16 – 26. 4 µm, light yellow to golden yellow, subglobose to ovoid or pyriform, echinulate, apical wall thickened, 1.5 im, germ pores one to two equatorial. Pedicles very short often not attached, wall thick. Telia and teliospores not found.

Specimen examined – Parasitic on *Canna indica* L cultivated, Bhanimandal, Lalitpur, Nepal. 2015.09.17 (2072. 05. 31) Adhikari, no. 207231. KATH

**Distribution**

Hawaii, South Africa, India, Thailand and Nepal

**Comments** – Telia and teliospores were not observed on any of the collected samples. This rust was not
found during extensive plant disease surveys in the south Pacific in the 1970s and early 1980s, but it was found in Fiji in 1984 and later in other South Pacific countries. Most of the Pacific collections have only uredinia, but one collection from Fiji has telia, and they have also been found in Hawaii. Pathogen identity was confirmed as *Puccinia thaliae* Dietel by nuclear ribosomal large subunit (28S) DNA sequencing with rust-specific primers (Kaur, Rush, Ferrin & Aime, 2011).

**References**


Dietel, P. 1899 *Uredinea brasilienses*, a cl. E. Ule lectae- II. *Hedwiglia*. 38:248-259


Nelson, S. 2013 Rust of Canna lily. *Plant disease*


Photograph

A. Photographs showing infected Canna

B. Close up view of rust

C. Urediniospores (10 x 40 magnification)
Effect of Different Soil Combinations on the Growth Rate of Ardisia macrocarpa Wall.

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Abstract

Ardisia macrocarpa Wall. commonly known as “Damai phal” is distributed in Nepal ranging from 1500-2400m above sea level. The present research work aimed to determine the suitable soil condition for the growth of A. macrocarpa. Study was carried out in green house at National Botanical Garden (NBG), Godawari, Lalitpur. Soil samples included forest, ground and sandy soil with different concentration of compost manure. Phenotypic traits such as height of plant, number of leaves and number of branches were recorded and used for data analysis. Morphological data showed range of variations in growing pattern of A. macrocarpa. The result showed that plant height, number of leaves and the number of branches were found to be maximum in the neutral sandy soil with pH of 6.20. The findings suggested that A. macrocarpa has a potential to attain growth under stress of nutrient availability.

Key words: Soil samples, phenotypic traits, growth rate, Ardisia macrocarpa.

Introduction

The study of phenological aspects of plant involves the observation, recording and interpretation of the timing of their life history events (Fenner, 1998). Natural population of plant shows intricate pattern of variation (Briggs and Walters, 1997). Intra and inter population variation in nature are nearly of quantitative rather than discontinuous kind (Falconer, 1981). Plants compete for different resources (eg; nutrients, space, light, moisture, etc). Only those survive, reproduce and increase in number which is best suited at that particular environmental condition (Morris et al. 2005). Plant grows on soil and their growth depends on quantity and quality of nutrients in it (Sharma, 2006). Soil type influences growth of the plant directly or indirectly by interacting with a large number of factors (Garg and Kumar, 2012).

The genus Ardisia belongs to the family Myrsinaceae. It consists of about 500 species worldwide. Three species of Ardisia occur in Nepal. The study species i.e. Ardisia macrocarpa Wall. is a shrub upto 2m high occurring in Central and Eastern Nepal at altitude ranging from 1500-2400m. In Nepali it is called as “Damai phal” (GoN, 2003). The leaves are 5-17cm long and 1.5-4cm broad, alternate, short petioled, lanceolate, narrowed at both ends, crisped crenulate with marginal row of pink dots and glabrous. Flowers are small, bisexual, stalked and white in colour. Corymb is sub-terminal. Fruit is a berry which is 0.7cm in diameter, globose, depressed, bright red and spotted. The flowering is in July and fruiting period is from October to January. It commonly occurs on shady forest floor (Shakya, 1986). Ripe berries are eaten raw especially by the people of Tamang community (Shrestha, 1988). A. macrocarpa is an evergreen shrub with attractive fruits. So, it was selected for the study to promote it in floriculture with its ornamental value. Research work on the growth rate of Ardisia is not yet performed in Nepal as well as other countries. This study aims to know the best soil condition for the growth of A. macrocarpa.

Material and method

Preparation of soil sample

Different soil samples such as ground soil collected from barren land, forest soil collected from the National Botanical Garden (NBG), Godawari, and sandy soil (sand mixed with soil in the ratio of 1:3) were prepared. The initial readings on pH and nutrients viz. Nitrogen (N), Phosphorus (P) and
Potassium (K) of each soil sample were measured. Various soil concentration of each soil samples were made by mixing it with compost manure in the ratios of 0:1 (neutral), 1:2, 1:3 and 1:4 compost manure to soil.

Seeds were collected from Conservation and Educational Garden of NBG in December 2013 and preserved in refrigerator at 18°C. Healthy seeds of *A. macrocarpa* were chosen and sown at a time in each combination of soil in January 2014. Data of vegetative characters like length of shoots and number of leaves were recorded frequently at an interval of 7 days from germination period. SPSS version 17.0 was used to perform t-test and graphs were prepared in MS Excel.

**Result and Discussion**

In Table 1, variability in soil pH and the basic plant nutrient N, P and K in all the studied soil samples at the initial period of the experiment are shown. The ground and forest soil were alkaline in nature whereas sandy soil was slightly acidic in nature. However, presence of N, P and K were high in forest soil whereas least in sandy soil.

At different composition of ground soil with compost manure, the average plant height, number of leaves and number of branches were found to be higher at 4:1 concentration (Figure 1). The maximum plant height was 26.4 cm, and the number of leaves was 39 while the number of branches was 3.

In the present study, Figure 2 shows the average increase in plant height, number of leaves and number of branches in sandy soil combinations of different concentration. The maximum plant height was 44.8 cm, number of leaves was 93, and the number of branches was 10 at 3:1 forest soil composition. Few plants grown in the pots containing this soil type died because the roots were eaten by insect called ‘khumre’ in nepali.

![Figure 1: Variation in the average plant height (Pl.Hgt.), number of leaves (Lf. no.) and number of branches (Br. no.) in different combinations of ground soil with compost manure.](image)

![Figure 2: Variation in the average plant height (Pl.Hgt.), number of leaves (Lf. no.) and number of branches (Br. no.) in different combinations of forest soil with compost manure.](image)

![Figure 3: Variation in the average plant height (Pl.Hgt.), number of leaves (Lf. no.) and number of branches (Br. no.) in different combinations of sandy soil with compost manure.](image)

<table>
<thead>
<tr>
<th>Soil type</th>
<th>pH(1:2.5% H₂O)</th>
<th>Total Nitrogen (N %)</th>
<th>Available Phosphorus (P₂O₅) Kg/ha</th>
<th>Available Potassium (K₂O) Kg/ha</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ground soil</td>
<td>7.60</td>
<td>0.23</td>
<td>34.09</td>
<td>482.4</td>
</tr>
<tr>
<td>Forest soil</td>
<td>7.60</td>
<td>0.25</td>
<td>45.10</td>
<td>616.4</td>
</tr>
<tr>
<td>Sandy soil</td>
<td>6.20</td>
<td>0.15</td>
<td>22.03</td>
<td>227.8</td>
</tr>
</tbody>
</table>
soil combinations of different concentration. The maximum plant height was 48.5 cm, the number of leaves was 138 and the number of branches was 15 at neutral sandy soil composition.

**Discussion**

Various factors such as soil pH, soil moisture, soil temperature, availability of micro and macro nutrients, etc. determine the growth and development of a plant (Korner, 2003). Growth rate of any plant is influenced by nutrient availability, its environment and their genetic constitution (Joshi and Joshi, 1998; Parmar and Pant, 2015). Macronutrients such as nitrogen and phosphorus required for the plant are mostly available at neutral soil pH (6-8) (Singh et al., 2006).

According to the findings of Muller and Brandes (1997) increase in shoot length of *Artemisia annua* in sandy soil was less than its normal height which is totally different from the result of this study. However, Nepali et al., (2015) carried similar work in *Lilium nepalense* found that sandy soil with less pH than other experimental soil samples has high growth of shoots, leaves and number of leaves. Similarly, the sand mixed with gravel supported the maximum increase in plant height, fresh weight and dry weight of *Euphorbia lathyris* followed by sand and other soils in Rajasthan (Garg and Kumar, 2012). Jackson (1987) also found that sandy-loam to sandy soil was the most suitable soil condition for the growth of *Dalbergia sissoo*. Sandy soil holds less moisture and nutrients per unit volume but permits more rapid percolation of precipitation water than other soils. From these above research works, it is clear that some plants prefer sandy soil for their proper growth. *A. macrocarpa* showed better growth in nutrient deficient soil than in nutrient rich soil which indicates that it has a potential to attain growth under stress of nutrient availability. The plant height, leaf number and branch number of *A. macrocarpa* were highest in neutral sandy soil which may be because it prefers slightly acidic soil.

**Conclusion**

From this study, plant height, number of leaves as well as number of branches were found to be higher in neutral sandy soil in comparison to forest soil and ground soil. Thus, it can be interpreted that *Ardisia macrocarpa* prefers sandy soil with pH of 6.20 for the propagation and better growth. Environmental factors or gene are responsible for the morphological characters. For the confirmatory test, it is suggested to progeny test, reciprocal transplant practice as well as study on molecular level.

**Acknowledgements**

We express our sincere gratitude to Mr. Rajdev Prasad Yadav, Director General of Department of Plant Resources, Mr. Dinesh Baral, Garden Officer and all the staffs of National Botanical Garden, Godawari.

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report submitted to NAST (Nepal Academy of Science and Technology).


Population status of *Curculigo orchoides* Gaertn. in a Community Managed Forest of Banke District, Nepal

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Abstract

The rhizomatous *Curculigo orchoides* Gaertn. (family Hypoxidaceae) is a medicinal plant with trade value in Nepal. Population structure of *C. orchoides* was studied in *Shorea robusta* and *Terminalia alata* forest Jalandhara Community Forest, Banke district, Western Nepal. It was abundantly found in the study forest with 87% frequency. This might be due to better management of the forest. Community user group of this forest can get benefit by the sustainable harvesting of this species.

**Key words**: Medicinal plant, Kalomusli, Population structure, Utilization, Management

Introduction

*Curculigo orchoides* Gaertn., belongs to family Hypoxidaceae, is commonly known as Kalomusli, Banjari (Tamang) and Musaleri (Tharu). The genus *Curculigo* comprises 70 species (www.tropicos.org). Out of 70 species, four species such as *C. capitulata*, *C. crassifolia*, *C. gracilis* and *C. orchoides* are found in Nepal (Press et al., 2005). Among them, *C. orchoides* is an important medicinal plant. The rhizome is used for demulcent, diuretic, tonic, aphrodisiac, piles, jaundice, asthma, diarrhoea and gonorrhrea (Anonymous, 2007). It is also one of the major ingredients of many Ayurvedic products like Aswagandharishta, Ghandak Rasayan (SDVKVS, 1999) and therapeutic products like Musale Churna, Shaktibardhak Yog, Tentex royal (Shrestha & Shrestha, 2004). Dried rhizomes are used for preparation of pharmaceutical drug *Rhizoma Curculiginis* (ESON, 2009).

Due to its wide range of uses in ethnomedicine, both in the Ayurvedic and Chinese systems of traditional health care, a large number of phytochemical and pharmacological investigations have been undertaken (Shrestha et al., 2008). Only few biological study of this plant has been done in Nepal. Phenology, population structure, and regeneration strategies of *Curculigo orchoides*, was studied in the inner Terai, Central Nepal by Shrestha et al. (2011).

People collect whole rhizome inhibiting vegetative propagation and thus affecting regeneration. Besides these other activities like land use changes, habitat destruction, uprooting and destroying entire plant during collection are considered major threats to this plant. Low regenerative potential through sexual reproduction and high vulnerability to habitat disturbance appear to be the major constraints for maintaining natural population of *C. orchoides* (Shrestha et al., 2011). Though wild population of *C. orchoides* are reported to be declining rapidly, and the plant has been assigned to various threat categories, as vulnerable (CAMP, 2001) and threatened (Manandhar, 2002), there is no detailed study on conservation status of this plant. Thus this study was undertaken to know its population structure in natural growing habitat.

Material and methods

**Study area**

The study site was selected by discussion with staffs of District Plant Resource Office and District Forest Office, Banke district. According to them, the availability of *Curculigo orchoides* was high in Jalandhara Community Forest of Banke district (Figure 1). The Jalandhara community forest (28°14’29” N Latitude and 81°76’76” E Longitude) is situated in Banke district, Bheri zone of Mid-
Western Development region. This district covers an area 2,337 km². Forests account for 50.17 per cent (0.11 million ha) of total land of the district. Major ecosystems in the district are Sal (*Shorea robusta*) forest, deciduous riverine forest, savannahs and grasslands, mixed hardwood forest, flood plain community, Bhabar and foot hills of Chure. The area of Jalandhara community forest is about 76 hectare and elevation ranges from 135 to 150 m.

To estimate the population of *C. orchioide* frequency (%), density, abundance was calculated (Zobel et al., 1987). Global Positioning System (GPS Garmin e Trex 10) was used to record the latitude, longitude and elevation for each plot.

The frequency was determined with the help of the following formula:

\[
\text{Frequency(%) = } \frac{\text{No. of plots with species}}{\text{Total no. of plots taken}} \times 100
\]

Density was calculated as:

\[
\text{Density (individual/ha) = } \frac{\text{Total no. of species} \times 10,000\text{m}^2}{\text{Total no. of plots} \times \text{size of quadrats}}
\]

After calculating frequency, density of the species, abundance was scaled up (Zobel et al., 1987).

Some secondary data about trade of this species were also collected from Jadibuti Association of Nepal (JABAN, 2014).

**Results and discussion**

During the present study, *C. orchioide* was found abundantly in Jalandhar Community Forest, of Banke district. The frequency of *C. orchioide* was low (57%) in sunny place and high (87%) in shady and marshy places. Similarly density of the species was minimum (11053 plant/hectare) and high (50869 plant/hectare) in sunny and shady place, respectively (Table 1). In sunny place, common dominant species associated with *C. orchioide* in studied populations were *Parthenium hysterophorus* L., whereas in shady place, we found mainly the associated species are *Murraya koenigii* (L.) Spreng., *Mallotus philippensis* (Lam.) Muell.-Arg. and grasses (Table 1). Field observation revealed that shady marshy open places with tree vegetation are the preferred habitat for this species. Shrestha et al. (2011) also reported that partial canopy are suitable for vegetative growth as well as flowering and fruiting of *C. orchioide*.

High frequency and plant density show that species preferred to grow in shady place. Low density in sunny place may be due to poor regeneration, low
seed germination and competition with invasive alien species *Parthenium hysterophorus*.

High frequency and density of the plant in the study site may be due to different reasons like suitable habitat and climatic condition for the plant, no anthropogenic disturbance and the better management of the forest. *C. orchioides* is listed as endangered and going to loss in natural habitat (CAMP, 2001). Although the species is categorized as endangered, there is no management plan for conservation in Nepal.

## Conclusion

It is concluded that the population of *Curculigo orchioides* in Jalandhara Community Forest is abundant. Sustainable harvesting, domestication and cultivation of such useful medicinal *C. orchioides* should be encouraged to fulfill market demand, which will increase the income of local people. Such economically important *C. orchioides* should be conserved with both in situ and ex situ methods of conservation. Further studies should be initiated to increase vegetative propagation, seed germination, seedling establishment and detail ecological adaptation of species to strengthen conservation program.

## Acknowledgements

We are thankful to Mr. Rajdev Prasad Yadav, Director General, Ms. Sushma Upadhyaya, Deputy Director General, Mr. Sanjeev Kumar Rai, Deputy Director General Department of Plant Resources, Thapathali, Kathmandu, for their kind support and encouragement. We are also grateful to, Mr. Arjun Neupane, Ms. Chandru Sharma, District Plant Resource Officers and field staffs of District Plant Resource Office, Banke, District Forest Office of Banke district for their help during field visit.

## References


<table>
<thead>
<tr>
<th>Collection site</th>
<th>Frequency</th>
<th>Density (individual/ha)</th>
<th>Abundance</th>
<th>Phenology</th>
<th>Associated plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sunny place</td>
<td>57.14%</td>
<td>11052.63</td>
<td>Frequent</td>
<td>No flowering</td>
<td><em>Parthenium hysterophorus</em>, <em>Murraya koenigii</em>, <em>Mallotus philippinensis</em>, grasses</td>
</tr>
<tr>
<td>Shady place</td>
<td>86.95%</td>
<td>50869.56</td>
<td>Dominant</td>
<td>Flowering</td>
<td><em>Murraya koenigii</em>, <em>Sida acuta</em>, <em>Mallotus philippinensis</em>, <em>Terminalia alata</em>, <em>Dioscorea bulbifera</em>,</td>
</tr>
</tbody>
</table>

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<th>Frequency</th>
<th>Density (individual/ha)</th>
<th>Abundance</th>
<th>Phenology</th>
<th>Associated plants</th>
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</thead>
</table>


Population Structure and Regeneration Status of Cyathea (Cyatheaceae) in Nepal

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Abstract

The tree fern (Cyathea) is distinct from other ferns by having erect trunk like stem. Tree fern is distributed throughout tropical and subtropical regions in central and eastern Nepal. Five species of tree ferns have been recorded from Nepal. The tree ferns is included in appendix-II of CITES list although there is no trade record. This paper identifies the localities where the tree fern is found and their regeneration status in central and eastern Nepal. The present study focused on field survey which was conducted between June to July, 2015 in selected VDCs of Kaski and Tanahu districts in central Nepal, secondary information collection and compilation as well as the views of district forest officers of the districts in Central and Eastern Nepal. The tree fern is found in 19 districts of Nepal. A total of 176 individual trees with 101 matured tree, 40 immature and 35 young stage were recorded with their geographical coordinates in Kaski and Tanahu districts. The occurrence of individuals indicates that the regeneration of tree fern is fair. Felling of trunk of Cyathea is major problem in central Nepal.

Key words: Cyathea, regeneration, Kaski district, Tanahu district

Introduction

Population structure and regeneration studies of species help to record the number of particular taxa in the definite natural areas. These types of research seem to be very indispensable to quantify individual species within the natural boundary of the country. Population and regeneration status, distribution and ecological characters of the plants provide information to conserve the species and encourage its sustainable use (Bhattacharya and Sharma, 2008).

Cyathea, the tree ferns of family Cyatheaceae are distinct from other ferns by having trunk like erect stem (Dong, 2009), some of them reaching upto 20 m in height (Tryon and Tryon, 1982), and includes about 600 species globally (Korall et al., 2006). The Cyathea is distributed from sea level to tropical and subtropical areas (Tryon and Gastony, 1975) in the world.

Nepal is well known for its unique topography and climatic conditions that provided suitable habitat for diversity of the flora and fauna. Fern diversity in Nepal is high and 550 species and 30 subspecies of ferns and fern allies are reported (Fraser-Jenkins et al., 2015). Altogether, six species of Cyathea namely Cyathea brunoniana, C. spinulosa, C. gigantea, C. henryi, C. khasyana and C. sollyana are documented mainly from central and eastern Nepal.

The genus Cyathea prefers to grow in moist habitats and is mostly terrestrial in nature. Among them Cyathea spinulosa is well known species for their excellent beauty of outdoor decoration. So, it is also raised in the gardens for ornamental purposes (Gurung, 1991). Local communities use young floral buds as food. In rainy days, the leaves are cut and used to make umbrellas. The trunk of the tree is extensively used to make pillar of traditional houses and shed. Local people in central Nepal consider that the trunk of Cyathea is as strong as the cemented pillar. In eastern India, the trunk of Cyathea species is exploited commercially while Cyathea gigantea trunk is popularly used to grow orchids (Khan et al., 2002). Leaves of the species are used to cure bodyache by the Apatani tribe (Kala, 2005); rhizome in combination with black pepper and milk are used to cure white discharge (Rout et al., 2009) as well as various decorative items like pots, flower vases, ash trays etc. are also prepared from the woody stem (Paul et al., 2015)
The species from genus *Cyathea* are included in CITES lists (www.cites.org). The species was previously considered as “vulnerable” in Nepal, and, more recently, as *Least concern (LC)* in 2001 in Nepal based on a CAMP workshop held in January 2001. There is no intensive study of the *Cyathea* with respect to its regeneration and distribution in Nepal. However, few literatures are available regarding the occurrence of the species (Gurung, 1991; Thapa, 2002; Bhattarai *et al.* 2004; Bhuju and Joshi, 2009; Bhagwat and Shrestha, 2010; Sharma *et al.*, 2013; Fraser-Jenkins *et al.* 2015) in different localities of country but no quantitative information including population structure of *Cyathea* is available for Nepal. No detailed study on *Cyathea* species has been carried out and very little information is available on population structure and regeneration status of the species. So, understanding the current population structure and regeneration status of *Cyathea* species is very important to develop an effective conservation initiative in the natural habitat.

The present study focuses on the study *Cyathea* regarding its population status, distribution and regeneration in central and eastern Nepal. The specific objective of the study are i) to assess the distribution of *Cyathea* species in central and eastern Nepal, ii) to assess the population structure of *Cyathea* species in some parts of central Nepal, and iii) to find out the regeneration status of *Cyathea* in its natural habitat.

**Taxonomy of Cyathea species found in Nepal**

*Cyathea* is the type genus of the family Cyatheaceae. They are mostly terrestrial ferns, usually with a single tall stem. Rarely, the trunk may be branched or creeping. Many species also develop a fibrous mass of roots at the base of the trunk. Taxonomic characteristics of *Cyathea* species recorded from Nepal according to Fraser-Jenkins *et al.*, 2015 is as follows

*Cyathea spinulosa* Wall. ex Hook.

The commonest tall tree fern reached up to of 4m height. The leaves of *C. spinulosa* are 2.5 to 6m long, bipinnate-tripinnatifid mostly spiny at the base and arranged spirally at the apex of the stipe with oblong, acute, serrulate segments. Stipe and rachis pale brown, prominently spiny, with smaller spines on the pinna-costae, stipe-bases bearing many narrowly lanceolate, bicolorus scale with darkish brown centre and thin pale toothed margins; pinnule costules and lobe midribs paler beneath, bearing small, whitish, lanceolate and deciduous scales beneath when young, becoming glabrous, but no hairs (below); young sori are surrounded by a thin white indusium, which shrivels and usually drops off on maturity. In Nepal, it occurs in warm and humid places at altitudes between 335 and 2000m (Gurung, 1991; Fraser-Jenkins *et al.*, 2015)

*Cyathea brunoniana* (C.B. Clarke) C.B. Clarke and Baker

A tall tree fern, trunk up to 6m height, very similar to *C. spinulosa* but it is found slightly in higher altitude, apex pale to mid brown, but stipe and rachis, though spiny, less so than *C. spinulosa* and with slightly smaller spines becoming warts further up the axes, diagnostically as well as scattered scales. It has small rather dense hairs clothing the undersurface of the costules or pinnule axes. It has sori with a small basal indusium at one side.

*Cyathea gigantea* (Wall. ex Hook.) Holttum.

A matured tree reaching up to height of 5m, stipe dark chestnut brown to nearly blace, matt, without spines, bearing many narrow chestnut-brown glossy scales with paler, fimbriate edge; rachis similar to other *Cyathea* but with few or no scales; frond bipinnate, tripinnatifid, ultimate lobes usually rather shallow. Fertile pinnules with markedly V-shaped rows of exindusiate sori.

*Cyathea henryi* (Baker) Copel.

This species is very much similar to that of *C. gigantea*. It only differs from *C. gigantea* by having longer and larger pinnules with more scales beneath their costules, more pairs of veinlets in the ultimate lobes, which are longer, becoming slightly rectangular and the lower ones on larger pinnae are more deeply separated from each other. The sori are found well apart forming weak V-shape but more parallel line than *C. gigantea*.
Cyathea khasyana (T. Moore ex Kuhn) Domin

One of the large tree fern reached up to the height of 4.7m. It is different from other Cyathea species by having non spiny stipe and rachis, minutely muricate below dark brown to purple brown, with a line of yellow stipes up the sides, bearing many narrow mid-casaneous brown, glossy scales towards the stipe-base with paler, ciliate margins, pinnule-lobes rather small, narrow and usually parallel sided, with small apical and often upper marginal teeth bearing many small dark-tipped, sori exindusiate.

Cyathea sollyana (Griff.) Fraser-Jenk.

The tallest tree fern of Indo-Himalaya reaching up to 15m height, usually an unbranched trunk, stipe and rachis pale to mid-brown, smooth but often slightly warty in the lower parts, glossy without spines or thorns, stipe-base scales linear-lanceolate, pale brown with darker cilia at the margins. White scales present beneath when leaf very young, fertile segments normally narrowed, completely covered in exindusiate sori; sori close packed along the midrib.

Materials and Methods

Study area

The study was carried out in selected Village Development Committees (VDCs) of Kaski and Tanahu districts of Central Nepal. Geographically Kaski district lies at 83°40' to 84°12' East longitudes and 28°06 to 28°36' North latitudes. Four types of climatic zones are found in these districts, subtropical, temperate, alpine and tundra climate. The total annual rainfall of Kaski districts is about 4200mm; maximum rainfall up to 1700mm occurring during August and similarly, the maximum temperature is recorded 32°C in summer and 2.2°C in winter season. Bhadaure Tamagi, Kande, Pumdibhumdi, Lwang-ghalel VDCs and Lekhanath municipality towards forested gully on east of Begnas lake were selected as sample area for field study mainly based on the availability of Cyathea species from Kaski district. Tanahu is adjoing district of Kaski and climatic condition is almost similar to that of Kaski. The Kahun Shivapur and Dorphirdi VDCs of Tanahu district were taken for sample study site.

Population structure and field Survey

Field survey for population structure and regeneration study of Cyathea species was conducted between June to July, 2015 in selected VDCs of each districts. Location of each of the individual Cyathea species was marked with GPS (Garmin 62cx). Field survey and data collection was based on the guidelines given by CITES COP-16.

A group discussion was conducted in the respective VDCs of the districts in order to generate accurate field level data of tree fern and their availability. The questionnaire was mainly focused on distribution of the species, regeneration status, part(s) used and occurrence of the species in the area. On the basis of villagers information the author with field assistance travelled to the forest area more than 5km each day in order to quote the species and their geo-coordinates. The walking distance was considered as the transect line and numbers of the tree ferns available both sides were recorded with coordinates, number of tree ferns available, their maturity stage as matured tree, immature and young stage trees were also recorded.

The individuals of Cyathea species recorded in the field were categorized into young individuals having upto 1.0m height, immature individuals having 1.0-1.5m height and mature individuals having more than 2.0m stem height.

To collect the information about the population structure of Cyathea from eastern Nepal secondary information mostly literatures were consulted. Besides secondary information from literatures and 39 district forest officer’s form mountain and Himalayan regions of central and eastern Nepal were consulted about the Cyathea species by phone calls. All the respective district forest officers took more than 10 days to collect the information and share with author. Other than Kaski and Tanahu district, the occurrence of Cyathea species based on the observation of respective district forest officers and their staffs in the field were recorded.
Results and Discussion

Distribution of Cyathea species

The distribution of Cyathea species is restricted to some parts of the 19 districts of eastern and central Nepal (Table-1). The dominance of Cyathea species is recorded from an altitude of 1,400m - 1,600m asl in the study site, though the plant showed distributed from 500m - 1,800m in Nepal. The Cyathea species in Pingshree, Shivanagar VDC in Tanahu district is confined mostly above 600 - 800m asl but that in Kaski district above from 1,400m asl. The present study found that the species is more frequently occurring at 1,400-1,600m asl in Kaski district of central Nepal along the bank of gullies, rivulets and permanent water sources. The genus Cyathea is mostly recorded from the bank of rivers, gullies and in shady, well-moistened places under the dark canopy of Alnus forest canopy in Lwangghalel VDC of Kaski district, however under the canopy of Schima wallichii-Castanopsis indica forest in Pingshree khola of Tanahu district.

The total numbers of Cyathea plant in the study site with mature tree, immature and young with their geographical coordinates are presented in Table-2. The Cyathea species is mostly associated with Schima wallichii, Castanopsis indica, Myrica esculenta, Engelhartia spicata, Erythrina arborescense in Pingshree Khola in Tanahu but it is associated mostly by Alnus nepalensis in Lwangghalel VDC of Kaski.

The present study recorded a total of 176 individuals of Cyathea species from Kaski and Tanahu districts with 101 individual matured trees, 40 of immature and 35 of young stage. Among them large number of trees were recorded in Lwangghalel VDC of Kaski having 89 trees with 50, 22 and 17 individuals of matured tree, immature and young stage respectively. The present study only covers about 10km of transect walk along the forest and gullies of Lwangghalel VDC. According to local communities the species mainly found in the present survey areas there have seen significant number of individuals also in nearby Kaskikot, Dhampus VDCs also. Similarly, 9 individual Cyathea species were recorded from Raniban with 5 at matured stage, 1 immature stage and 3 at young stages and 26 individual from Deurali-Tamagi VDC of Kaski where all individuals

Table 1: Cyathea recorded from districts and their location

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Districts</th>
<th>Areas within the districts</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Manang</td>
<td>Jagat-Dharapani</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>2</td>
<td>Palpa</td>
<td>Ridikhola</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>3</td>
<td>Parbat</td>
<td>Panchase Area</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>4</td>
<td>Kaski</td>
<td>Near Pokhara, Dhampus, Begnas, Panchase, Lwangghalel, Kande,</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>5</td>
<td>Tanahu</td>
<td>Kahunshivapur-2,pingsirikhola, Near khairenitar-Chhabise village</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>6</td>
<td>Gorkha</td>
<td>Komale and Balu khola</td>
<td>On shady &amp; moist places</td>
</tr>
<tr>
<td>7</td>
<td>Lamjung</td>
<td>Opposite side of Marsyandi river near Jagat/Syang/ Ghermu VDC-5, 6, Tagrin VDCs</td>
<td>More than 100 large tree</td>
</tr>
<tr>
<td>8</td>
<td>Dhading</td>
<td>Raniban below Nagarjun ridge below Jamachok</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>Kathmandu</td>
<td>Sundarijal</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Bhaktapur</td>
<td>Nagarkot, Sankhu near Changunarayan temple</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>Dolakha</td>
<td>Totalabari, Tamba-Bhotekoshi valley</td>
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</tr>
<tr>
<td>12</td>
<td>Solukhumbu</td>
<td>Dudhkoshi</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>Sankhuwasava</td>
<td>Near Arunriver below Num, Arun valley</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>Bhojpur</td>
<td>Dingla, Chirkhowa</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>Ilam</td>
<td>Rangapani, satpokhari, Pyang VDC 1850m asl</td>
<td>Krishna Ram Bhattarai, posted on facebook</td>
</tr>
<tr>
<td>16</td>
<td>Panchthar</td>
<td>On the way to Phidim (12 pokhari)</td>
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</tr>
<tr>
<td>17</td>
<td>Taplejung</td>
<td>Garhi Danra-Linkim-Tuwa, Tamur valley, mewa khola</td>
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<tr>
<td>18</td>
<td>Nuwakot</td>
<td>Different VDCs of northern belt</td>
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<td>19</td>
<td>Kavre</td>
<td>Some places of borders sites towards Bhaktapur</td>
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Table 2: Areas surveyed under study showing geographical positions and number of trees recorded

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<tr>
<th>S.N.</th>
<th>Village/Area</th>
<th>Altitude m</th>
<th>Latitude</th>
<th>Longitude</th>
<th>No. of Matured tree</th>
<th>No. of immature tree</th>
<th>No. of young</th>
<th>Estimated no. of tree fern in 10x10 m plot</th>
<th>Remarks</th>
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<td>28.1966</td>
<td>83.9661</td>
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<td>1</td>
<td>1</td>
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<td>3m apart</td>
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<td>28.1916</td>
<td>83.9665</td>
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<td>0</td>
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<td><strong>9</strong></td>
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<td>1627</td>
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<td>2</td>
<td>3 WGS 84</td>
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<td>4km west from Deurali</td>
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</tbody>
</table>

Conclusion: A total of 53 individuals of *Cyathea* were recorded from Pingshree khola of Shivanagar VDC of Tanahu district with 32 individual at matured tree, 11 and 10 individuals of immature and young stage respectively (Table 2).
Regeneration status of Cyathea

Regeneration status of the species was calculated on the basis of relative proportion of individuals at young, immature and mature stage. From the present study it is revealed that in all samples VDC the number of mature tree is higher than in young and immature. Paul et al. 2015 showed that 60% population regeneration of C. gigantea in eastern Himalaya (India) is at very poor regeneration stage. This result is more or less similar to the present study. However, the immature and young stage tree ferns are present in all studied sites indicating that the regeneration rate is slow but showed either fair or good or poor regeneration condition in Lwangghalel, Bhadaure Tamagi, Rani ban and Pingshree Khola of Tanahu district (Table 1)

Conclusion

Present study found a total of 176 Cyathea individual trees with 101 at matured, 40 at immature and 35 at young stage recorded from Kaski and Tanahu districts. About 63% of the observed individuals were matured tree and only 20% individual were at young stage indicating that the population regeneration is fair. Similarly the Cyathea species is recorded from 19 districts of eastern and central Nepal. The felling of tree trunk to make the pillar of local houses and shed is a challenging problem to conserve the species in Nepal.

Acknowledgements

The author is thankful to Mr. Rajdev Yadav (Director General, DPR), Ms. Sushma upadhaya and Mr. Sanjeev K. Rai, Deputy Director Generals of DPR for their valuable inspiration and providing working environment. Author strongly acknowledges Prof. Dr. Ram Prasad Chaudhary for valuable suggestions and guidance and Ms. Sangeet Swar (Head of Biodiversity Section) for field work as well as setting working situation during the field work with local people of Kaski and Tanahu districts. The author is specially thankful to Mr. Rabindra Budha, Mr. Kalyan Sapkota, Mr. Sushil Lamsal and Ms. Saroja Adhikari for their help during field work.

References


[www.cites.org/eng/resources/species.html](http://www.cites.org/eng/resources/species.html)
Documentation of Ethnomedicinal Knowledge on Plant Resources Used by Baram Community in Arupokhari VDC, Gorkha District, Central Nepal

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Abstract

The present paper highlights 80 species of plants used as medicine by the Baram (Baramu) community of Arupokhari VDC of Gorkha District. These reported plants were used in cough, cold, fever and gastritis, wound, diarrhea and other diseases.

Keywords: Ethno-medicine, Baram community, Arupokhari VDC, Gorkha district

Introduction

Ethno-botanical studies required to document and explore the traditional knowledge hidden in different ethnic group (Manandhar, 1993, Pandey, 2013, Parajuli, 2013). Among the 103 indigenous groups of Nepal, Baram (Baramu) is also one such ethnic group. Baram (or Baramu) is one of the indigenous minority nationalities scattered in different villages in the districts of Gorkha and territories (CBS, 2011). According to their mythology, they call themselves off-springs of what they call five brothers: Surel, Sunuwar, Jirel, Rai (Khambu) and Limbu, and claim to have scattered to the west from the east (Gautam and Thapa Magar, 1994). The Baram people are a Tibeto-Burman ethnic group (Kansakar et al, 2011). The objective of this study is to document the traditional knowledge of Baram community regarding ethno-medicine.

Materials and Methods

This study was conducted in indigenous Baram community at Arupokhari VDC, Gorkha district, Central Nepal during September 2015. This VDC inhibits most common Mountain Sal forest, Chirpine forest and Schima-Castanopsis forest at altitude ranges from 500 m asl to 1300 m asl. In the course of study, 8 experienced villagers (5 male and 3 female) were interviewed of age range between 40-80 years old. The plants were identified with the help of photographs and related literatures (Polunin and Stainton, 1984, Baral and Kurmi, 2006, Jha et al., 2008, Manandhar, 2002, Shrestha, 1998, Shrestha and Shrestha, 2004, Stainton, 1998).

Results and Discussion

Total 80 species of 74 genera belongs to 44 families being used by Baram community as medicine to cure 27 diseases and troubles. Some major families are Moraceae (7 spp.), Asteraceae (6 spp.), Fabaceae (6 spp.), Lamiaceae (4 spp.), Verbenaceae (4 spp.) and others used to cure different diseases. On the basis of life form (habit) of plants 29 herb, 16 shrub, 29 tree and 6 climber species being used from past. Among all, 14 species being using in cough, 9 species in cold, 9 species fever, 9 species gastritis, 8 species in wound, 7 species in diarrhea and so on.

Ethno-medicinal use of plants

The information is arranged in alphabetical order of botanical name followed by family, nepali name, roman name habit and uses.

**Abrus precatorius** L., Fabaceae, रतिगेडी / लालगेडी, Ratigedi, Climber

Root directly is used to reduce hotness in stomach due to eating *Capsicum* spp.

**Acorus calamus** L., Araceae, ब्होजो, Bhojo, Herb

Juice of rhizome is drink to cure cough, cold and diarrhea.
**Agave americana** L., Agavaceae, कैल्याण / केंद्रीक, Kettuke, Shrub
Leaves juice is apply to kill worms on wound.

**Ageratum conyzoides** L., Asteraceae, गंधे, Gandhe, Herb
Juice of leaves is applied to stop bleeding in cut.

**Ageretina adenophora** (Spreng.) King & H.Rob., Asteraceae, बनमारा, Banmara, Herb
Juice of leaves is applied to stop bleeding.

**Allium sativum** L., Amaryllidaceae, लसुं, Lasun, Herb
Bulb is directly eating to reduce gastritis.

**Aloe vera** (L.) Burm. f., Liliaceae, ग्हुकुमरा, Ghiy Kumara, Herb
Juice of leaves is applied to cure skin burn by fire.

**Ananas comosus** (L.) Merr., Bromeliaceae, भूलफुल, Bhui katahar, Herb
Fruit is directly eating to reduce warmness of body.

**Artemisia indica** Willd., Asteraceae, तिते, Tite pati, Herb
Juice of leaves is applied to kill human louse, worms and also cure scabies.

**Artocarpus heterophyllus** Lam., Moraceae, कठर, Katahar, Tree
Leaves is directly fed cattles to control cough.

**Artocarpus lacoocha** Wall. ex Roxb., Moraceae, बडहार, Badahar, Tree
Milky latex is applied to cure mumps.

**Azadirachta indica** A. Juss., Meliaceae, नीम, Neem, Herb
Juice of leaves is applied to kill human louse, worms and also cure scabies.

**Bauhinia variegata** L., Fabaceae, कौरी / कोइरालो, Koiralo, Tree
Juice of bark is eating to cure fever, diarrhea as well as amebic dysentery.

**Belamcanda chinensis** (L.) Redoute, Iridaceae, तंबारे फूल / बुधनी धार, Tarbare phool/ Khukuri dhare, Herb
Decoction of root is eating to cure diarrhea.

**Boeninghausenia albiflora** (Hook.) Reichenb. ex Meissn., Rutaceae, माँगे माउर / माउरे भार, Makhe Mauro/ Maure jhar, Herb
Plant decoction is eating to cure cold and whole plant used for insect repelent.

**Bombax ceiba** L., Bombacaceae, सिन्हल, Simal, Tree
Dried flower is directly eating to cure dysentery.

**Buddleja paniculata** Wall., Buddlejaceae, भिमसेन पाती, Vimsen pati, Tree
Juice of leaves is use to kill fishes.

**Callicarpa arborea** Roxb., Verbenaceae, गूयालो, Guyalo, Tree
Decoction of bark is eating to cure fever.

**Callicarpa macrophylla** Vahl., Verbenaceae, दही चामल / दहीकंकला, Dahichamle, Shrub
Decoction of bark as well as root is eating to cure fever, typhoid.

**Celtotropis gigantea** (L.) Dryand., Asclepiadaceae, आक, Aank, Herb
Milky juice is applied to cure joint fracture and dried stem is use as smoke in Pinas.

**Cannabis sativa** L., Cannabaceae, गाँजा, Ganja, Herb
Decoction of leaves is fed to cure diarrhea of goat and other cattles.

**Centella asiatica** (L.) Urb., Apiaceae, घोड़ताप्प, Ghodtapre, Herb
Decoction of plant is eating to cure fever and to reduce warmness of body.

**Cheilanthes bicolor** (Forssk.) Kaulf., Pteridaceae, रानी सिंंका / काली सिंंका, Ranisinka, Herb
Mixture of leaf decoction of *Scutellaria discolor*, *Psidium guajava* and *Cheilanthes bicolor* is eating twice a day to cure gastritis troubles.

**Chromolaena odorata** (L.) King & H.E. Robins., Asteraceae, बनमारा / बनमारा, Banmara, Shrub
Decoction of leaves is applied to stop bleeding as well as skin ring of cattle.

**Cissampelos pariera** L., Menispermaceae, गुजर गानो, Gujar gano, Climber
Decoction of rhizome /root is eating to cure gastritis problems.

**Citrus aurantifolia** (Christ.) Swingle, Rutaceae, कागड / कागड़, Kagati, Lemon, Tree
Juice of fruit is applied to cure pimple.

**Clustocalyx operculata** (Roxb.) Merr. & Perry, Myrtaceae, ब्यामुनो, Kyamuno, Tree
Dry leaves is used as smoke to cure cold, Pinas. *Colebrookea oppositifolia* Sm., Lamiaceae, धुसुरी, Dhusure, Shrub

Juice of young bud is applied to remove leech from nose of cattle.

*Costus speciosus* (Koenig) Sm., Zingiberaceae, बेतलौरी, Betlauri, Herb

Juice of root is eating to cure fever.

*Costus speciosus* (Koenig) Sm., Zingiberaceae, बेतलौरी, Betlauri, Herb

Juice of root is eating to cure fever.

*Lyonia ovalifolia* (Wall.) Drude, Ericaceae, अंगेरी, Angeri, Tree

Juice of leaves/bud is applied to control scabies.

*M smallest* (Wall.) DC., Myrsinaceae, बाङे, Bango, Shrubs

Juice of leaves is used to fish poisoning.

*Milletia extensa* Benth., Fabaceae, गाउजो, Gaujo, Shrub

Juice of root is applied to control scabies.

*Mimosa pudica* L., Fabaceae, लाजवाती, Lajjawati, Herb

Juice of leaves is eating to cure jaundice, reduce warmness of body.

*Morus alba* L., Moraceae, किम्बु, Kimbu, Tree

Latex is used in anthe-helmintic for cattle.

*Musa paradisiaca* L., Musaceae, केरा, Kera, Herb

Juice of young bud of flower is eating to cure jaundice.

*Mussaenda macrophylla* Wall., Rubiaceae, अविश्वी, Avijalo, Herb

Juice of root is eating to cure fever.

Myrica esculenta Buch.-Ham. ex D. Don, Myricaceae, ररराफल, Kaphal, Tree

Juice of leaves applied to control toothache.

*Nicotiana tabacum* L., Solanaceae, सुरी, Surti, Herb

Juice of leaves applied to kill and remove leeches from the nose of cattle.

*Nyctanthes arbor-tristis* L., Oleaceae, नायरताल, Parijat, Shrub

Juice of leaves is drink to cure cough.

*Ocimum sanctum* L., Lamiaceae, तुलसी, Tulas, Herb

Leaves boil in water and drink to cure fever, cough, cold.

*Oroxylum indicum* (L.) Kurz, Bignoniaceae, टतेलो, Tatelo, Tree

Decoction of bark is drink to cure jaundice, seed eat in fever and paste of bark applied in wound.

*Osbeckia nepalensis* Hook., Melastomaceae, सेतो, Seto chulsi, Shrub

Leaves boils in water and drink to cure fever.
Phyllanthus emblica L., Euphorbiaceae, Amala, Tree
Fruit is eat to cure cold and cough.

Pinus roxburghii Sarg., Pinaceae, Khote salla, Tree
Resin is applied to cure mumps.

Piper longum L., Piperaceae, Pipla, Climber
Fruit is directly eating to cure cold and cough.

Pogostemon benghalensis (Brum. f.) Kuntze, Lamiaceae, Rudilo, Herb
Decoction of leaves use to cure cold and cough.

Premna latifolia L., Verbenaceae, Ginneri, Tree
Juice of bark is drink to reduce gastritis problem.

Prunus persica (L.) Batsch., Rosaceae, Aaru, Tree
Juice of young bud is applied to kill worms on wound.

Psidium guavaja L., Myrtaceae, Amba, Tree
Juice mixture of leaves of Psidium guavaja, Scutellaria discolor and Cheilanthes bicolor is drink twice a day to cure gastritis and Bark decoction use to cure diarrhea.

Rhododendron arboreum Sm., Ericaceae, Lalligurans, Tree
Flower is directly eating to control diarrhea.

Rubus ellipticus Sm., Rosaceae, Aiselu, Shrub
Juice of root is drink to cure gastric and young bud used in stomach pain.

Saccharum spontaneum L., Poaceae, Kans, Herb
Root paste is directly applied to remove helminths of cattle.

Sapium inisgne (Royle) Benth.ex Hook.f., Euphorbiaceae, Khirro, Tree
Milky latex is used in fish poisoning.

Schima wallichii (DC.) Korth., Theaceae, Chilaune, Tree
Juice of bark is used in fish poisoning as well as drink in gastritis problem.

Scutellaria discolor Colebr., Lamiaceae, Ratopate, Herb
Juice mixture of leaves of Scutellaria discolor, Psidium guavaja and Cheilanthes bicolor is drink twice a day to cure gastritis. Root decoction is drink to cure fever.

Solanum virginiatum L., Solanaceae, Kanti kari, Herb
Dry fruit is used as smoke in toothache.

Spermacoce latifolia Aubl., Rubiaceae, Kune jhar, Herb
Juice of whole plant is applied to cure wound.

Spilanthes paniculata Wall.ex DC., Asteraceae, Marauti, Herb
Flower is applied to control toothache and stomach pain due to cold.

Stephania glandulifera Miers, Menispermaceae, Batulpate, Climber
Root juice is drink to cure cough.

Syzygium cumini (L.) Skeels, Myrtaceae, Jamuna, Tree
Juice of bark is drink to cure dysentery.

Tectaria macrodonta (Fee) C.Chr., Aspidiaceae, Kali niuro, Herb
Root juice is drink to cure dysentery.

Terminalia bellirica (Gaertn.) Roxb., Combretaceae, Barro, Tree
Fruit is directly eating to cure cold and cough.

Terminalia chebula Retz., Combretaceae, Harro, Tree
Fruit is directly eating to cure cold and cough.

Urtica dioica L., Urticaceae, Sisno, Herb
Leaves paste is use to make bandage in joint fracture/breakage.

Vitex negundo L., Verbenaceae, Simali, Shrub
Juice of leaves is used to cure cough and Pinas.

Woodfordia fruticosa (L.) Kurz, Lythraceae, Dhayero, Shrub
Flower is directly eating to cure dysentery.

Zanthoxylum armatum DC., Rutaceae, Timur, Shrub
Fruit is directly applied to cure toothache.

Zingiber officinale (Willd.) Roscoe, Zingiberaceae, Aduwa, Herb
Rhizome is directly eat to cure cold and cough.

Herbal plants are used in the form of juice, decoction, paste and also use directly by chewing. The most common cold and cough problem were treated by
using some plants like *Phyllanthus emblica*, *Pogostemon benghalensis*, *Terminalia bellirica*, *Terminalia chebula*, *Zingiber officinale*, *Acorus calamus* and *Ocimum sanctum*. Similarly, *Cheilanthes bicolor*, *Allium sativum*, *Cissampelos pariera*, *Osbeckia nepalensis*, *Premna latifolia*, *Scutellaria discolor* and *Rubus ellipticus* were used to cure gastritis. Most of the milky latex bearing plants (*Artocarpus lakoocha*, *Ficus benghalensis*, *Ficus racemosa*, *Ficus sarmentosa*) used to cure mumps problem. Only two species *Mussaenda macrophylla* and *Callicarpa macrophylla* are used to treat typhoid disease.

**Conclusion**

The Baram people of Gorkha district Central Nepal possess rich ethno-medicinal knowledge and practiced several species of plants to cure different diseases and health troubles. The globalization and modernization creating problem to conserve the Baram language as well as traditional practice to use medicinal plants. Documentation as well as bioprospecting and patenting is very important to provide fair share of benefit to indigenous people.

**Acknowledgements**

We are grateful to Plant Resources Officer, N.H. Chapagain, District Plant Resources Office, Makwanpur for his encouragement on research activities. Our sincere thanks go to local people (especially Ran B. Baram) of Arupokhari VDC, Gorkha District for their participation and kind cooperation during the field study.

**References**


Documentation of Indigenous Knowledge on Medicinal Use of Plants by Raji Community in West Nepal

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Abstract
The present work was carried out with the aim to document the indigenous knowledge on medicinal use of plants by Raji community in west Nepal. The study was mainly based on primary data collected during the explorative field study representing locality with significant Raji population. As per the approved annual program of District Plant Resources Office (DPRO) Kailali for the fiscal year 2014/15, the work was carried out in the year 2015. Among 61 plant species were identified in the present study highest number of species was recorded in Bhuruwa, Khailad-4 of Kailali. Similarly, highest number belongs to herbs (22 species) followed by trees (20 species) of trees. The most commonly used part of plants for medicinal purpose was root (24 species) followed by leaf (14 species). According to types of health problems, greater numbers of plant species (12 species) are found to be used by Raji to treat gastrointestinal problem.

The study revealed that Raji community still has a strong traditional knowledge on medicinal plants use and they are still applying traditional healing practices. But strength of indigenous knowledge and practice is higher in the locality where Raji population is in significant number with their own socio-cultural settings. The strength becomes weaker and weaker with decrease in households and population along with socio-cultural erosion.

Key Words: Indigenous knowledge, Raji, plants, medicinal uses, west Nepal.

Introduction
Indigenous knowledge is such type of empirical idea and methodology, which is generated through series of hits and trials, from generations to generations. In other words, indigenous Knowledge is human life experience in distinct natural and social compound within unique local and contemporary setting. It is not formally taught but perceived in particular context at a certain stage of the perceiver’s consciousness that grows in the world of local events (Maskay, 2007). Indigenous knowledge or synonymously traditional knowledge is tested and refined knowledge selected through thousands of years or even more.

Nepal is not only rich in biodiversity but also rich in ethnic diversity. Each ethnic community has localized distribution with unique socio-cultural characteristics. Raji is an indigenous community and are believed to originate from Surkhet with most households today living in the Chure hills and low land Terai of the Mid Western Region and the Far Western Region. The Rajis speak a Tibeto-Burman language without a script. The Rajis are divided into three clans the Purbe (Atharathari), the Bandale (Barathari) and the Naukale (Nauthari). The language also varies slightly according to clan.

According to National Census 2011, there is only 4235 Raji population in Nepal. Raji are mainly distributed in Mid Western and Far Western regions and represented by 2106 and 2036 in number respectively. Eastern, Middle and Western regions have sparse distribution of Raji and represented by significantly low numbers 33, 30 and 30 respectively (CBS, 2011). The largest Raji population is found in Surkhet and Kailali districts, followed by Dang, Bardiya and Kanchanpur districts. Government of Nepal (GoN) has enlisted the Raji community in vulnerable community list. According to Social Protection Programme Implementation Guidelines
(GoN, 2008), each Raji is entitled to NRs. 1000 per month from the respective VDC or municipality office under the Social Protection Programme of the Government of Nepal.

The Rajis used to live a nomadic life in the past (Gautam & Thapa Magar, 1994) and prefer to live along river banks and within forest areas. However, with the adoption of a semi-nomadic lifestyle, they tend to gradually move out of the forest. Agriculture is a newly embraced occupation for Raji and probably the last among other communities, which tends to rely on it for their self-subsistence, although most Raji families also remain dependant on traditional means of survival. Raji are renowned for their skills at honey hunting and fishing. Amongst the Raji a specific classification of diseases and concepts prevail and although the Rajis have begun to use modern medicine, they generally resort to it only their own traditional healers fail to cure the disease, making it important for them to retain their knowledge in this regard (UNRCHC, 2012).

Statement of the problem

Many indigenous communities have been abandoning their traditional customs and thereby lose their plant knowledge over time (Benzet et al., 2000). Change in life style, urbanization, ignorance of new generation and biodiversity loss are major causes of significant decrease in traditional knowledge on medicinal plants among various ethnic communities of Nepal (Maskey, 2007; Thapa, 2012). Indigenous Knowledge is traditionally transmitted orally but is now vulnerable to people’s migration and the youth adopting new and different lifestyles and values. The Rajis’ substantial Indigenous Knowledge is also at risk of disappearing with the older generation of Rajis.

Objectives

Following are the objectives of present study;
- To document the indigenous medicinal practices by Raji community.
- To analyze the results of research.

Methodology

Literature review was first activity conducted to carry out the research. Various relevant documents were collected and studied. On the basis of literature review, methodology and study site was finalized.

Study Area

Since the research is mainly confined with traditional medicinal plant uses by Raji community, the study areas are selected where Raji community lives from generations to generation. Accordingly following are the study areas.

<table>
<thead>
<tr>
<th>Study site</th>
<th>District</th>
<th>Geographical Position</th>
<th>Altitude</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Krishnapur – 5</td>
<td>28° 50.602' N; 80° 28.636' E</td>
<td>196 m</td>
</tr>
<tr>
<td>2</td>
<td>Bhuruwa, Khailad VDC – 4</td>
<td>28° 29.717' N; 80° 55.539' E</td>
<td>147 m</td>
</tr>
<tr>
<td>3</td>
<td>Chhinchu – 11</td>
<td>28° 27.869' N; 81° 43.009' E</td>
<td>559 m</td>
</tr>
<tr>
<td>4</td>
<td>Kuta, Taranga – 4</td>
<td>28° 41.313' N; 81° 23.862' E</td>
<td>325 m</td>
</tr>
<tr>
<td>5</td>
<td>Sanashree, Taratal MN– 2</td>
<td>28° 41.313' N; 81° 23.862' E</td>
<td>325 m</td>
</tr>
</tbody>
</table>

Research Design and Data Collection

Exploratory research design was selected for present study. The study mainly focuses on collecting and analyzing the first hand information. While, secondary data were also analyzed at the discussion part of the study. Elders, traditional healers, farmers...
of the community were identified as key informants from preliminary discussion. Field study was conducted in 2015.

Interview method was used to collect the primary information about medicinal uses of plants. Semi-structured open-ended questionnaire was used while interviewing the respondent. But before taking the interview, prior informed consent was taken from each and every respondent. After completing the interview, transect walk was conducted in the nearby forest where information was added and herbarium specimens were collected for uncommon species. Audio recordings were also taken in the interview and in the transect walk, and the recording were used in verifying the data. The specimens were identified with the help of standard literatures and technical supports from experts of KATH. The herbarium prepared during the study was stored in DPRO Kailali Herbarium.

Results and Discussions

Number of medicinal plant species used by Raji in different locality

![Figure 1. Number of medicinal plant species used by Raji in different localities](image)

In the study, highest number of species was recorded in Bhuruwa, Khailad of Kailali, where 30 species of medicinal plants were found to be used to treat different health problems. This figure was followed by Taranga and Chhinchu of Surkhet where 27 and 26 species of medicinal plants were recorded respectively. Krishnapur, Kanchanpur stands in 4th position with seven medicinal plants used by Raji community. Lowest number of species of medicinal plant was reported in Taratal of Bardiya. The research team could not make contact with majority of key informants at Taratal, because at the time of field study, most of the people of the community were engaged in public contribution program for local development work, popularly called as ‘begari’.

Taxonomic Diversity

Altogether 61 plant species were identified which are used by Raji community for medicinal purpose. Among these, 46 species were identified up to species level, 5 species up to genera and three species upto family level only while remaining 8 were unidentified because the specimens were not at flowering or fruiting stage. Among identified species belongs to 29 family and 46 genera. During the study, 42 species belonging to 26 families of dicotyledons, 3 species belonging to 2 families and 2 genera of Monocotyledon, one species of Pteridophytes were recorded. The largest family was Asteraceae having 7 species followed by Fabaceae (4 spp.), Apocynaceae (3 spp.), Combretaceae (3 spp.), Phyllanthaceae (3 spp.), Rutaceae (3 spp.).

Life forms

![Figure 2. Life forms of medicinal plant species used by Raji](image)

Based on life forms, highest number belongs to herbs (22 species, 34.42%) followed by trees (20 species, 32.79%) of trees, shrubs (10 species, 16.39%), climbers (7 species, 11.49%). Lowest number is represented by woody climber with 3 species (4.92%) which are found to be used by Raji community for medicinal purpose.
Plant parts

![Bar chart showing the most commonly used parts of plants for medicinal purposes](image)

The most commonly used part of plants for medicinal purpose was root (24 species, 39.34%) followed by leaf (14 species, 22.95%), bark (12 species, 19.67%), fruit (7 species, 11.48%), young shoot and seed both (4 species, 6.56%). 6 species (9.84%) have other parts such as whole plant, leaf sap, leaf latex, stem, bulb, flower etc. used for medicinal purpose. Some species have more than one part used for medicinal purpose; the virtual number of species seems to be more than total number of species enumerated in the study.

Treatment of health problem category

![Bar chart showing the number of species used to treat health problems by Raji](image)

According to types of health problems, greater number of plant species (12 species, 19.67%) are found to be used by Raji to treat gastrointestinal problem, which is followed by gradually lesser number of species of medicinal plants to treat ENT problems (10 species, 16.39%), wounds and external damages (9 species, 14.75%), heart, liver kidney problems (7 species, 11.48%), bone fractures (6 species, 9.84%), cattle health problems (5 species, 8.20%) and antidote against snake and scorpion bites (3 species, 4.92%). Remaining species are used to treat against fever, muscle pain, headache, arthritis etc.

Conclusion

Altogether 61 species of medicinal plants are found to be used by Raji to treat the health problems. From above results, it can be concluded that Raji community still have a strong traditional knowledge on medicinal plants use and they are still applying traditional healing practices. But strength of indigenous knowledge and practice is higher in the locality where Raji population is in significant number with their own socio-cultural settings. The strength becomes weaker and weaker with decrease in households and population along with socio-cultural erosion. The high diversity of medicinal plant species used by Raji of Bhuruwa, Khailad-4 of Kailali is closely linked with the fact that Bhuruwa is most likely the highest number of households with highest population of Raji in Nepal. The finding is supported by greater number of medicinal plant species reported from Chhinchu and Taranga followed by lower number of medicinal plant species used by Raji in Kanchanpur and Bardiya.

Acknowledgements

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References


### Appendix

List of plant species and their information collected during Traditional Knowledge of Raji of West Nepal

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Scientific Name</th>
<th>Family</th>
<th>Habitat</th>
<th>Raji Name</th>
<th>Nepali/Local Name</th>
<th>Medicinal Uses</th>
<th>Parts Used</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Achyranthes aspera L.</td>
<td>Amaranthaceae</td>
<td>Herb</td>
<td>Chichibhrata</td>
<td>in abdominal pain (abdominal ache), for body freshness</td>
<td>Roots, Leaves</td>
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<tr>
<td>2</td>
<td>Ageratum conyzoides L.</td>
<td>Asteraceae</td>
<td>Herb</td>
<td>Raunia</td>
<td>in cuts and wounds</td>
<td>Leaves</td>
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<td>3</td>
<td>Alstonia scholaris (L.) R. Br.</td>
<td>Apocynaceae</td>
<td>Tree</td>
<td>-</td>
<td>in abdominal disorders, gastritics</td>
<td>Barks</td>
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<td>4</td>
<td>Antidesma acidum Retz.</td>
<td>Phyllanthaceae</td>
<td>Tree</td>
<td>Chau</td>
<td>in wounds</td>
<td>Leaves</td>
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<tr>
<td>5</td>
<td>Artemisia sp.</td>
<td>Asteraceae</td>
<td>Shrub</td>
<td>Pati</td>
<td>in short urination period</td>
<td>Roots</td>
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<td>6</td>
<td>Asparagus flicinus Buch.-Ham. ex D. Don</td>
<td>Asparagaceae</td>
<td>Herb</td>
<td>Bhale Gansya</td>
<td>in fractures of limbs</td>
<td>Roots</td>
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<td>7</td>
<td>Asparagus racemosus Wild.</td>
<td>Asparagaceae</td>
<td>Herb</td>
<td>Pothi Gansya</td>
<td>in fractures of limbs, wounds, cuts, body pain, for the increment on the prolactine hormone and hence the mother's milk</td>
<td>Roots</td>
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<td>8</td>
<td>Bergenia ciliata Stemb.</td>
<td>Saxifragaceae</td>
<td>Herb</td>
<td>Pasanved</td>
<td>in naval rigidity, abdominal pain</td>
<td>Roots</td>
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<td>9</td>
<td>Bridelia retusa (L.) A.Juss</td>
<td>Phyllanthaceae</td>
<td>Tree</td>
<td>Drek</td>
<td>headache, body pain</td>
<td>Seeds, Barks, Roots</td>
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<td>10</td>
<td>Calotropis gigantea (L.) R. Br. ex Schult.</td>
<td>Apocynaceae</td>
<td>Shrub</td>
<td>Madar</td>
<td>in the rabies, sutzure and pain in vessels</td>
<td>Flower, Leaves</td>
<td></td>
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<td>11</td>
<td>Carica papaya L.</td>
<td>Caricaceae</td>
<td>Tree</td>
<td>Mewa</td>
<td>in stinged wounds of Centipede, in different stones, spermatorrhea</td>
<td>Leaves, Seeds/Roots</td>
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<td>12</td>
<td>Cassia fistula L.</td>
<td>Fabaceae</td>
<td>Tree</td>
<td>Aainrokha</td>
<td>in the snakebites</td>
<td>Leaves ans stem tip</td>
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<td>13</td>
<td>Centella asiatica (L.) Urban</td>
<td>Apiaceae</td>
<td>Herb</td>
<td>Ghodtapre</td>
<td>in black spot tongue disease, to escape from high temperature</td>
<td>Leaves/Whole plant</td>
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<td>14</td>
<td>Cissampelos pareira L.</td>
<td>Menispermaceae</td>
<td>Shrub</td>
<td>Ganogurjo</td>
<td>in naval rigidity of large mammals, oxes</td>
<td>Underground stem</td>
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<tr>
<td>15</td>
<td>Cissampelos sp.</td>
<td>Menispermaceae</td>
<td>Climber</td>
<td>Khalfya</td>
<td>in diarrhoea, abdominalache, naval rigidity</td>
<td>Roots</td>
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<td>16</td>
<td>Clausena pentaphylla DC.</td>
<td>Rutaceae</td>
<td>Shrub</td>
<td>Pradha</td>
<td>in abdominalache</td>
<td>Roots</td>
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<td>17</td>
<td>Cleistocalyx operculatus (Roxb.) Merr. &amp; L.M. Perry</td>
<td>Mirtaceae</td>
<td>Tree</td>
<td>Bhadrajabu</td>
<td>in sinusitis, cuts and wounds</td>
<td>Leaves</td>
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<tr>
<td>18</td>
<td>Clematis sp.</td>
<td>Ranunculaceae</td>
<td>Climber</td>
<td>Shikari lahara</td>
<td>in fractures of limbs</td>
<td>Roots</td>
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<tr>
<td>19</td>
<td>Colebrookea oppositifolia Sm.</td>
<td>Lamiales</td>
<td>Shrub</td>
<td>Dhurseul</td>
<td>In conjunctivitis, and eye diseases</td>
<td>Newly tips</td>
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<td>20</td>
<td>Cynoglossum sp.</td>
<td>Boraginaceae</td>
<td>Herb</td>
<td>Kaniike kuro</td>
<td>In conjunctivitis, and eye diseases</td>
<td>Newly tips</td>
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<td>21</td>
<td>Datura metel L.</td>
<td>Solanaceae</td>
<td>Shrub</td>
<td>Dhaturo</td>
<td>in hydroseal</td>
<td>Leaves and Fruits</td>
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<td>22</td>
<td>Eclipta alba (L.) Hassk.</td>
<td>Ecliptaceae</td>
<td>Herb</td>
<td>Kali/Angare jhar</td>
<td>In filthy mud, skin corrosion</td>
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<td>Elephantopus scaber L.</td>
<td>Euphorbiaceae</td>
<td>Herb</td>
<td>-</td>
<td>in pathogens over the wounds of animals</td>
<td>Roots</td>
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<td>24</td>
<td>Ficus racemosa L.</td>
<td>Moraceae</td>
<td>Tree</td>
<td>Umri</td>
<td>in quick maturity of wounds</td>
<td>Latex of leaves</td>
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<tr>
<td>25</td>
<td>Ficus semicordata Buch.-Ham. ex Sm.</td>
<td>Moraceae</td>
<td>Tree</td>
<td>Khurure</td>
<td>in marasmus</td>
<td>Fruits</td>
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<td>Hydrocotyle sp.</td>
<td>Ranunculaceae</td>
<td>Herb</td>
<td>Binase jhar</td>
<td>in sinusitis</td>
<td>Leaves</td>
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<td>Lygodium flexuosum (L.) Sw.</td>
<td>Lygodiaceae</td>
<td>Herb</td>
<td>Kwanblaka</td>
<td>in dry cracks of foot base</td>
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<td>28</td>
<td>Millettia extensa (Bent h.) Baker</td>
<td>Fabaceae</td>
<td>Woody climber</td>
<td>Gaujo</td>
<td>in ticks and mites of cattle and in fishing</td>
<td>Roots</td>
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<td>No.</td>
<td>Plant Name</td>
<td>Family</td>
<td>Plant Type</td>
<td>Part(s) Used</td>
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<td>Momordica charantia L.</td>
<td>Cucurbitaceae</td>
<td>Climber</td>
<td>Karella, Karella</td>
<td>in controlling of blood pressure</td>
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<td>Murraya koenigii (L.) Sprengel</td>
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<td>Shrub</td>
<td>Daineri, Karipatta</td>
<td>lice, ticks and mites of cattle</td>
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<td>31</td>
<td>Oroxylum indicum(L.) Benth. ex Kurz</td>
<td>Bigoniaceae</td>
<td>Tree</td>
<td>Pradha, Tatelo</td>
<td>in conjunctivitis, vomit control, jaundice, abdominal swell, wounds and knee displacement</td>
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<td>Oxalis cornuculata L.</td>
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<td>Herb</td>
<td>Khapsurung, Chariamilo</td>
<td>in naval rigidity, conjunctivitis</td>
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<td>33</td>
<td>Phanera vahlii Benth.</td>
<td>Fabaceae</td>
<td>Woody climber</td>
<td>M ik lahara, Bhora</td>
<td>in abdominalache, normal bowel, dysentery</td>
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<td>34</td>
<td>Phyllanthus emblica L.</td>
<td>Phyllanthaceae</td>
<td>Tree</td>
<td>Aunila, A mala</td>
<td>in coughs</td>
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<td>Piper peepuloides Roxb.</td>
<td>Piperaceae</td>
<td>Climber</td>
<td>Pimpali, Pipala</td>
<td>in coughs</td>
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<td>36</td>
<td>Prunus persica (L.) Batsch</td>
<td>Rosaceae</td>
<td>Tree</td>
<td>A aru, A aru</td>
<td>in pathogens over the wounds of cattle</td>
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<td>37</td>
<td>Rauvolfia serpentina (L.) Benth. ex Kurz</td>
<td>Apocynaceae</td>
<td>Herb</td>
<td>Dhadbiruwa, Sarpagandha</td>
<td>in heart pain</td>
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<td>Rhus wallichi Hook.f.</td>
<td>Rutaceae</td>
<td>Tree</td>
<td>Ryakh, Valayo</td>
<td>in conjunctivitis and eye diseases</td>
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<td>39</td>
<td>Sapindus mukorossi Gaertn.</td>
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<td>Tree</td>
<td>- Ritha</td>
<td>in pains parts</td>
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<td>Schleichera oleosa (Lour.) Merr.</td>
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<td>- Kusum</td>
<td>in control of lices</td>
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<td>Smilax ovatifolia Roxb. ex D. Don</td>
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<td>Climber</td>
<td>Raiblu, chela, Kukurdino</td>
<td>in arthritis, joint pain</td>
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<td>Solanum nigrum L.</td>
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<td>Herb</td>
<td>Kakhani, Kaliyedi, Kaliyuni</td>
<td>in braco-clavical pain</td>
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<td>Shrub</td>
<td>Dudhi</td>
<td>in miscarriage</td>
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<td>44</td>
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<td>Woody climber</td>
<td>Sine, Moya, Debrelahara</td>
<td>in pain and sprains</td>
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<td>Spondias pinnata (L. f.) Kurz</td>
<td>Anacardiaceae</td>
<td>Tree</td>
<td>Amaro, Amaro</td>
<td>in abdominalache</td>
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<td>46</td>
<td>Terminalia arjuna (Roxb.) Wight &amp; Arn.</td>
<td>Combretaceae</td>
<td>Tree</td>
<td>Arjun, Arjun</td>
<td>in fractures of limbs</td>
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<td>47</td>
<td>Terminalia bellirica (Gaertn.) Roxb.</td>
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<td>Vayarang, Barro</td>
<td>in urinary track infection and urinary problems</td>
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<td>Terminalia chebula Retz.</td>
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<td>in coughs, dysentery, gingivitis, scurvy</td>
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<td>49</td>
<td>Triumpheta bartramia L.</td>
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<td>Herb</td>
<td>Biskapro</td>
<td>in wounds</td>
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<td>Vernonia aspera Buch. - Ham.</td>
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<td>Herb</td>
<td>Bareauns/ Thulouasadi</td>
<td>in abdominalache</td>
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<td>Ziziphus mauritiana Lam.</td>
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<td>in jaundice, toothache</td>
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<td>Asteraceae</td>
<td>Herb</td>
<td>Kalli, Kalijiri, Kalojira</td>
<td>in heart pain, cough, nausea, vomiting</td>
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<td>53</td>
<td>-</td>
<td>Herb</td>
<td>Bhule, Jangali besar</td>
<td>in cuts and wounds</td>
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<td>Bons</td>
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<td>Shrubs</td>
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<td>in irregular menstrual period</td>
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<td>56</td>
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<td>Tree</td>
<td>Simgane</td>
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<td>57</td>
<td>-</td>
<td>Tree</td>
<td>Byanki, Hadchur</td>
<td>in fractures, wounds, lymphy dots</td>
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<td>58</td>
<td>-</td>
<td>Herb</td>
<td>Choti, Banmula</td>
<td>in naval rigidity of oxes</td>
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<tr>
<td>59</td>
<td>-</td>
<td>Climber</td>
<td>Bhedko aauns, Bveddy</td>
<td>in wounds and cuts</td>
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<td>Fudune, Jhar</td>
<td>in pneumonia</td>
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<tr>
<td>61</td>
<td>-</td>
<td>Herb</td>
<td>Lodi</td>
<td>in conjunctivitis, and eye diseases</td>
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Photographs of some unidentified species
Screening of Physico – Chemical Parameters and chemical composition of essential oil of Turmeric Leaves (*Curcuma domestica*)

Ramesh K Yadav, Parasmani Yadav, Jyoti Joshi Bhatta and Dipesh Kattel  
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**Abstract**

The essential oil from dried leaves of *Curcuma domestica* (Zingiberaceae), commonly known as turmeric, was isolated by hydro-distillation using clevenger apparatus and the oil percentage was 1.0%. Specific Gravity, Refractive Index, Optical Rotation, Acid Value, Ester Value were determined and TLC was performed. The GC-MS analysis of the oil showed presence of terpenes, alcohols and some other compounds. The major compounds were Phellanderene (alpha), Terpinolene, Eucalyptol, Limonene, Myrcene, Pinene (alpha), Terpinene (gamma), Cymene (para), Terpinene (alpha), Pinene (beta), Carene (delta-3), Ocimene (E-beta), Terpen-4-ol.

**Key Words:** *Curcuma, leaves, essential oil, physico-chemical properties, chemical composition, GC-MS, TLC*

**Introduction**

*Curcuma domestica* (Zingiberaceae) is an annual herb commonly known as turmeric. The plant is indigenous to Bangladesh, Sri Lanka and India and is also widely cultivated in China, Japan, Brazil, Nepal and Thailand. It is selected as One Village One Product (OVOP) reference for Sunsari district by Nepal Government along with other twenty two products into different districts. Essential oils are complex mixtures of secondary plant metabolites which can be obtained from turmeric flowers, barks, roots, leaves, peels, seeds etc.

Physicochemical properties of oil like colour, odour, density, specific gravity, refractive index, optical rotation, acid value, etc indirectly influence the quality of essential oil. The commercial importance of oil mostly depends on these physicochemical properties, which provide baseline data to determine its suitability for consumption.

Turmeric leaf oil has various chemical compounds that include phellandrene, limonene, zingiberene, curcumene, turmerone and cineole.

The *Curcuma domestica* can be used for the prevention of Brain tumors, prostate cancer, skin cancer, leukemia, multiple myeloma, metastasis prevention, chemotherapy enhancer, Alzheimer’s prevention, eye health, weight loss, natural painkiller & anti-inflammatory, liver health, Parkinson’s disease, multiple sclerosis, depression, better sleep, antibacterial. It has long been used as a folk medicine and used to some extent as a digestive aid and in the treatment of fever, infections, dysentery, arthritis and other physiological problems. Some literature reviews showed that the Turmeric leaf oil can be used as biofuel in alternative of petrol.

**Materials and Methods**

Fresh leaves of *Curcuma domestica* were collected from Saptari district located in Eastern Development Region of Nepal in January 2015 and dried in shade for one month. Leaves (5Kg) were hydro distilled in a Clevenger apparatus for six hours. The yield of essential oil was 1.0%. The oil thus obtained was dried over anhydrous sodium sulfate and stored in a sealed glass vial at low temperature prior to analysis.
Physico – Chemical Properties

The values of Physico – Chemical Parameters determined were as follows:

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Parameters</th>
<th>Results</th>
<th>Methods</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>Colour</td>
<td>Pale Yellow</td>
<td>hydro-distillation of dried leaves using Clevenger apparatus, British Pharmacopia, Vol. 11.1988 (Appendix X I E A 137E volatile oil in Drug)</td>
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<tr>
<td>2</td>
<td>Odour</td>
<td>Characteristic</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Oil Percentage</td>
<td>1.0%</td>
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<tr>
<td>4</td>
<td>Specific Gravity</td>
<td>0.8555 at 24.6 ºC</td>
<td>A OAC 19th Edition, 2012 (Vol. II Ch-41, Page 2-3 method no. 985.19</td>
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<tr>
<td>5</td>
<td>Refractive Index</td>
<td>1.476 at 26.8 ºC</td>
<td>ISO 280:1999 (E)</td>
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<tr>
<td>7</td>
<td>Acid Value</td>
<td>0.203</td>
<td>ISO 1242:1999 (E)</td>
</tr>
<tr>
<td>8</td>
<td>Ester Value</td>
<td>8.8454</td>
<td>British Standard methods of tests for essential oils (1953)</td>
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</table>

Thin Layer Chromatography (TLC)

TLC was carried out in the solvent system of n-hexane:ethyl acetate (96:4). Anisaldehyde methanolic sulfuric acid (0.5ml Anisaldehyde + 10 ml Glacial acetic acid + 85 ml Methanol + 5 ml conc. Sulfuric acid) was used as developing reagent and TLC plate was heated in oven at 110 ºC for 10 minutes. Six Spots were observed with Rf values 0.11, 0.22, 0.30, 0.58, 0.78 and 0.95.

GC/MS Analysis

The oil obtained from hyrodistillation was injected on a GCMS – QP 2010 Plus (SHIMADZU), Gas Chromatography Mass Spectrometer instrument using a Rtx-5MS column (30m X 0.25mm X 0.25 µm), and with library NIST 11 and FFNCS 1.3. Carrier gas was Helium.

Fig.1: Showing the temp. program, chromatogram and chemical constituents of Curcuma domestica oil
Results and Discussion

The oil was obtained by conventional hydro distillation of the leaves of Curcuma domestica in a Clevenger apparatus. The oil percentage obtained was 1.0% on fresh weight basis. Compounds were identified by the GCMS instrument by making comparison with MS-library: Nist-11 and FFNSC 1.3. GC-MS analysis resulted in the identification of following compounds: Phellanderene (alpha) 28.61%, Terpinolene 19.64%, Eucalyptol 8.84%, Limonene 6.60%, Myrcene 5.89%, Pinene (alpha) 5.47%, Terpinene (gamma) 5.23%, Cymene (para) 4.22%, Terpinene (alpha) 3.73%, Pinene (beta) 2.40%, Carene (delta-3) 2.73%, Ocimene (E-beta) 3.03%, Terpen-4-ol 1.13%, p-menth-1-en-8-ol 0.96%, Sabinene 0.79% and Linalool 0.74%, in the turmeric leaves oil.

Physico – chemical parameters (Specific Gravity: 0.8555 at 24.6 pC, Refractive Index: 1.476 at 26.8 pC, Optical Rotation: 38.83p at 27.83 pC, Acid Value: 0.203 and Ester Value: 8.8454 were determined.

Conclusion

We conclude that the essential oil of turmeric leaves can be isolated by simple hydrodistillation method and thus the chemical composition and other physio-chemical parameters were studied. The findings of this study indicate the presence of the major chemical constituents as terpenes, Phellanderene (alpha), Terpinolene, Limonene, Myrcene, Pinene (alpha) and alcohol: Eucalyptol which is also consistent with number of spots obtained in TLC. The TLC performed of the oil can be henceforth used as reference TLC for further analysis.

Acknowledgements

The authors are grateful to the Director General of DPR Mr. Rajdev Prasad yadav, Deputy Director General Mrs. Susma Upadhyaya for their great motivation and Mr. Rajendra Sharma, Mr. Krishna Kumar Shah, Mr. Rajeswar Ranjitkar Mr. Keshab Poudel, Mr. Anjani Kumar Adhikari and Mr. Govinda Prasad Gautam are also thankful for their cooperation and encouragement.

References


Topographical variation of chemical constituents of Essential Oil of *Rhododendron anthopogon* (sunpati) leaves by Gas Chromatography-Mass Spectrometry

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Abstract
The chemical composition of essential oil of *Rhododendron anthopogon* leaves obtained from hydrodistillation process was investigated by using gas chromatography-mass spectrometry (GC-MS). The chemical constituents in essential oil may vary with soil conditions, altitudes, climatic conditions and other environmental factors. The essential oils obtained from the plant leaves collected from three different places of Nepal were thus analyzed by using GC-MS and compared. It was found that the major chemical constituents present in the oils of the given plant collected from the three different places varied to some extent.

Keywords: essential oil, *Rhododendron anthopogon*, GC-MS analysis, topographical variation.

Introduction

Essential oils are complex mixtures, constituted by terpenoid hydrocarbons, oxygenated terpenes and sesquiterpenes which originated from the plant secondary metabolism and are responsible for characteristic aroma of plant [1]. Essential oils are valuable natural products used as raw materials in many fields such as perfumes, cosmetics, aromatherapy, spices and nutrition [2]. *Rhododendron anthopogon* is an evergreen shrub growing up to an altitude of 4,500 m a.s.l. found in temperate zone of Nepal from east to west. This plant is commonly known as Anthopogon and in Nepal as Sunpati. Due to its aromatic properties, it is widely used as incense. Furthermore, Himalayan healer (or Amchi) uses its leaves and fresh flowers as tea for drinking to promote digestive heat, stimulate appetite and to relieve liver disorders. *R. anthopogon* is also used for sore throat, common cold and lung problems [3]. The essential oil of *R. anthopogon* known as anthopogon oil or sunpati oil (in Nepal) is commonly obtained by steam distillation of its aerial parts and is a good natural source of a faintly balsamic essence. Anthopogon oil can be used on the skin and hair [3]. According to Himalayan aromatherapy, this oil stimulates the nervous system and has been used for treating sore muscles and gouty rheumatic conditions [3]. The major components of the aerial parts of the oil are the monoterpenes â-pinene, â-pinene, limonene and the sesquiterpene â-cadinene [2]. By far the research conducted on major chemical constituents of an essential oil have shown that the several factors such as nutrients, environmental conditions, extraction processes, drying methods, soil conditions, climatic conditions, etc. affects the components in essential oils [4-8].

The development of gas chromatographic technique has facilitated the separation of volatile components in essential oil and mass spectrometry detector has provided a means to identify chemical compounds. Thus, in recent years a hyphenated system, GC-MS in which gas chromatography is coupled with mass spectrometry detector would initially separates a volatile organic mixture into its components by chromatography which are then further identified tentatively by mass spectrometry using a MS library.

Experimental Methods and Materials

a) Plant sample collection
Fresh leaves of *R. anthopogon* were collected from three different places of Nepal located at different
altitude in the same season. These plant samples were collected from Lauribina of Gosakunda area, central region of Nepal, Jiri of Dolakha District, central region and Papung of Talpejung district, eastern region of Nepal located at an altitude of about 4300m, 3700m and 2000m a.s.l. respectively. These plant samples were then identified at Natural Products Research Laboratory (NPRL), Kathmandu and were left to dry in shade for some weeks.

b) Extraction of essential oil from leaves
100gm leaves of each sample of *R. anthopogon* collected from the three different places were subjected to the hydrodistillation process using Clevenger apparatus to extraction the essential oil for about 5 hours. The percentage of essential oils i.e. oil % obtained from each 100gm sample was also noted. The oils from each three places thus obtained were separated from the hydrosol, tagged and then stored at 4°C for further analysis.

c) Analysis of essential oil samples by GC-MS
The chemical constituents in the essential oils were separated using a Shimadzu gas chromatography (GC 2010) with Rtx-5MS column (25m×0.25mm×0.25µm). 0.5 µL of undiluted essential oil was injected into the GC inlet maintaining column flow rate of 0.8 mL/min and purge flow 3 mL/min after fixing the split ratio at 90.0. The initial column oven temperature was set at 50.0°C and the injection temperature was 180°C.

Table 1: Oven Temperature Program.

<table>
<thead>
<tr>
<th>Rate</th>
<th>Temperature (°C)</th>
<th>Hold Time (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>-</td>
<td>50.0</td>
<td>1.00</td>
</tr>
<tr>
<td>9.00</td>
<td>150.0</td>
<td>0.00</td>
</tr>
<tr>
<td>9.00</td>
<td>206.0</td>
<td>0.00</td>
</tr>
<tr>
<td>15.00</td>
<td>250.0</td>
<td>3.00</td>
</tr>
</tbody>
</table>

GCMS analysis for the qualification of the essential oil was carried out in a Shimadzu GCMS-QP2010 Plus. During the analysis, the ion source temperature and the interface temperature was set at 200°C and 250°C respectively. The detector scanning start time was 4.10 min and end time was 24.00 min; scan speed was 2500 with scanning range of m/z 40.00-1090.00. The MS library used in the analysis process was NIST 11, FFNSC 1.3.

Results and Discussion
*R. anthopogon* belongs to the Ericaceae family. Altogether 3 samples of the plant under study were collected from three different places of Nepal.

The oil percentages of *R. anthopogon* leaves obtained during hydrodistillation are tabulated below in Table 2.

Table 2: Oil % of Sunpati leaves samples of three different places.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Sample collected from</th>
<th>Oil percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Gosaikunda</td>
<td>0.5</td>
</tr>
<tr>
<td>2</td>
<td>Dolakha</td>
<td>0.5</td>
</tr>
<tr>
<td>3</td>
<td>Taplejung</td>
<td>0.4</td>
</tr>
</tbody>
</table>

The chromatograms obtained for each of the oil sample are shown below:
Fig 2: Chromatogram of anthopogon oil from Gosaikunda

Fig 3: Chromatogram of anthopogon oil from Dolakha

Fig 4: Chromatogram of anthopogon oil from Taplejung
Table 3: Chemical composition of essential oils of *R. anthopogon* leaves collected from three different places of Nepal

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Chemical Constituents</th>
<th>Oil from Gosaikunda</th>
<th>Oil from Dolakha</th>
<th>Oil from Taplejung</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Pinene &lt;alpha-&gt;</td>
<td>10.00</td>
<td>7.48</td>
<td>26.10</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Pinene &lt;beta-&gt;</td>
<td>5.23</td>
<td>7.36</td>
<td>13.59</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Cymene &lt;para-&gt;</td>
<td>2.06</td>
<td>-</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Limonene</td>
<td>2.09</td>
<td>1.13</td>
<td>9.03</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>beta Oicimene</td>
<td>0.82</td>
<td>4.04</td>
<td>1.89</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>Terpinene &lt;gamma-&gt;</td>
<td>5.25</td>
<td>4.98</td>
<td>3.08</td>
<td></td>
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<tr>
<td>7</td>
<td>2-Octanol, acetate</td>
<td>1.21</td>
<td>1.16</td>
<td>-</td>
<td></td>
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<td>8</td>
<td>Lilanol</td>
<td>1.38</td>
<td>-</td>
<td>1.37</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>p-Menth-1-en-8-ol</td>
<td>0.65</td>
<td>0.94</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Citronellyl acetate</td>
<td>4.14</td>
<td>3.88</td>
<td>0.59</td>
<td></td>
</tr>
<tr>
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<td>Copaene &lt;alpha-&gt;</td>
<td>0.95</td>
<td>0.90</td>
<td>1.02</td>
<td></td>
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<tr>
<td>12</td>
<td>Caryophyllene &lt;(E)-&gt;</td>
<td>12.52</td>
<td>11.55</td>
<td>4.97</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>Farnesene &lt;(E)-, beta-&gt;</td>
<td>5.2</td>
<td>4.39</td>
<td>2.37</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>Humulen &lt;alpha-&gt;</td>
<td>1.63</td>
<td>1.25</td>
<td>0.80</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>Isogermacrene D</td>
<td>3.27</td>
<td>3.94</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>16</td>
<td>Eudesma-4(14),11-diene</td>
<td>1.26</td>
<td>-</td>
<td>1.44</td>
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</tr>
<tr>
<td>17</td>
<td>Amorphene &lt;alpha-&gt;</td>
<td>3.84</td>
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<td>-</td>
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<tr>
<td>18</td>
<td>beta-Curcumene</td>
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<td>-</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>19</td>
<td>Cadinene &lt;gamma-&gt;</td>
<td>3.59</td>
<td>-</td>
<td>4.22</td>
<td></td>
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<tr>
<td>20</td>
<td>Cadinene &lt;delta-&gt;</td>
<td>6.34</td>
<td>6.61</td>
<td>9.50</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>alpha-M uroloene</td>
<td>0.42</td>
<td>-</td>
<td>7.88</td>
<td></td>
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<tr>
<td>22</td>
<td>Nerolidol &lt;(E)-&gt;</td>
<td>0.43</td>
<td>-</td>
<td>-</td>
<td></td>
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<tr>
<td>23</td>
<td>Citronelloyl valerate</td>
<td>0.94</td>
<td>0.83</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>Spathulenol</td>
<td>0.50</td>
<td>-</td>
<td>-</td>
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<tr>
<td>25</td>
<td>Caryophyllene oxide</td>
<td>3.01</td>
<td>3.14</td>
<td>-</td>
<td></td>
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<tr>
<td>26</td>
<td>Epiglobulol</td>
<td>0.73</td>
<td>-</td>
<td>4.22</td>
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<tr>
<td>27</td>
<td>Cubenol</td>
<td>0.41</td>
<td>-</td>
<td>-</td>
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<td>28</td>
<td>Cubenol &lt;1,10-di-epi-&gt;</td>
<td>1.71</td>
<td>1.72</td>
<td>-</td>
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<tr>
<td>29</td>
<td>Eudesmol &lt;epi-gamma-&gt;</td>
<td>1.25</td>
<td>1.11</td>
<td>-</td>
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<tr>
<td>30</td>
<td>Cadin-4-en-10-ol</td>
<td>16.59</td>
<td>8.63</td>
<td>1.37</td>
<td></td>
</tr>
<tr>
<td>31</td>
<td>Eicosane &lt;n-&gt;</td>
<td>1.35</td>
<td>-</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>32</td>
<td>M ycerene</td>
<td>-</td>
<td>3.23</td>
<td>2.82</td>
<td></td>
</tr>
<tr>
<td>33</td>
<td>o-Cymene</td>
<td>-</td>
<td>1.07</td>
<td>1.07</td>
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<tr>
<td>34</td>
<td>trans-alpha-Bergamotene</td>
<td>-</td>
<td>4.73</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>35</td>
<td>tau-M urolool</td>
<td>-</td>
<td>10.79</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>36</td>
<td>Nonadecane &lt;n-&gt;</td>
<td>-</td>
<td>1.34</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>37</td>
<td>Aromadendrene</td>
<td>-</td>
<td>-</td>
<td>1.01</td>
<td></td>
</tr>
<tr>
<td>38</td>
<td>Amorphene &lt;epsylon-&gt;</td>
<td>-</td>
<td>-</td>
<td>3.98</td>
<td></td>
</tr>
<tr>
<td>39</td>
<td>Cubene &lt;beta-&gt;</td>
<td>-</td>
<td>-</td>
<td>0.51</td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>(-)-delta-Cadinol</td>
<td>-</td>
<td>-</td>
<td>1.84</td>
<td></td>
</tr>
</tbody>
</table>

The above table shows the major components in the anthopogon oil are:

- Pinene <alpha->
- Pinene <beta->
- Limonene
- Terpinene <gamma->
- Citronellyl acetate
- Caryophyllene <(E)->
- Farnesene <(E)-, beta->
- Isogermacrene D
- Cadinene <gamma->
- Cadinene <delta->
- Cadin-4-en-10-ol
- Myrcene

Here, some major chemical constituents have more or less similar composition % in all the oil samples collected from the three different places. Except, in the oil sample from Taplejune which contain...
pinene<α->, pinene<β-> and limonene with higher % but citronellyl acetate, caryophyllene<(E)->, farnesene<(E)-,β-> and isogermae D have lower % compared to the oils from Gosaikunda and Dolakha. Further some chemical components present in the oil from Gosaikunda are absent in the oils from Dolakha and Taplejung, but it lacks the major component myrcene. The high % constituents: beta-ocimene, trans-alpha-bergamotene and tau-murolol present in oil from Dolakha are almost absent in the oils from Gosaikunda and Taplejung. Similarly, alpha-murolene, epiglobulol and amorphene <ε> that have high % in the oil from Taplejung are almost absent in the oils from Gosaikunda and Dolakha.

Conclusion

The present study shows that Oil % is slightly high in plant samples collected from Gosaikunda and Dolakha than from Taplejung. The major chemical constituents of the oils under study were Pinene<α->, Pinene<β->, Limonene, Terpinene<γ->, Citronellyl acetate, Caryophyllene<(E)->, Farnesene<(E)-,β->, Isogermae D, Cadinene<γ->, Cadinene<δ-> and Myrcene.

Further, the variation in chemical composition in oil samples of different places indicates that topography is one of the major factors affecting the chemical constituents of the essential oil of *Rhododendron anthopogon* (sunpati). These topographical differences can be considered to be the differences in soil conditions and altitudes, however since the sample collection were carried out in the same season and of random plants the climatic conditions and other factors are not studied. Thus, we can finally conclude the chemical constituents of essential oil of *Rhododendron anthopogon* vary due to the topographical variation.

Acknowledgements

The authors are highly obliged to Mr. Yam Bahadur Thapa (Former Director General, DPR) and Mrs. Sushma Upadhyaya (Deputy Director General, DPR), for providing us with the necessary laboratory facilities and resources. We would like to express our sincere thanks to Mr. Tara Datta Bhatta (Head of instrument section, DPR) and Mrs. Jyoti Joshi (Chief, NPRL) for their constructive suggestions and encouragement. And we would also like to acknowledge the entire family member of Department of Plant Resources, Thapathali, for their unflinching help and support.

References


Screening of the Physico – Chemical Parameters and chemical composition of essential oil of Citronella (Cymbopogon winterianus Jowitt)

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ramesh8276@gmail.com

Abstract

The essential oil of fresh leaves of Citronella (Cymbopogon winterianus Jowitt) was isolated by hydro-distillation method using clevenger apparatus and the oil percentage was 1.4 %. Specific Gravity, Refractive Index, Optical Rotation, Acid Value and Ester Value were determined and TLC was performed. In total, fifteen compounds were identified by GCMS analysis and some of these major compounds were Nerol, Neral, Nerol acetate, Cetronellol, Cetronellal.

Key Words: Citronella, Fresh leaves, essential oil, physico-chemical parameters, chemical composition, GC-MS, TLC

Introduction

Citronella (Cymbopogon winterianus Jowitt) is an aromatic grass belonging to the family Poaceae which gives essential oils upon steam distillation. It is a perennial herb which forms one-meter-high compact and strong clumps, with extensive use in popular medicine. This is used extensively as a source of perfumery, soap, cosmetic and flavoring industry throughout the world. It is used as antmyotic, acaricide and repellent against a variety of insects. Traditionally used for the treatment of fever, intestinal parasites and digestive problems. It can be used as massage oil for aching joints and muscles. The essential oils are natural products that exhibit a variety of biological properties, such as analgesic anticonvulsant and anxiolytic.

Materials and Methods

Fresh leaves of Citronella were collected from Dhanusha district located in Western Development Region of Nepal in May 2015. Collected fresh leaves were hydro distilled in a Clevenger apparatus for six hours. The yield of essential oil was 1.4%. The oil thus obtained was dried over anhydrous sodium sulphate and stored in a sealed glass vial at low temperature prior to analysis.

Physico – Chemical Parameters

The Physico – Chemical Parameters determined were as follows:

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Parameters</th>
<th>Results</th>
<th>Methods</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Colour</td>
<td>Light Yellow</td>
<td>hydro-distillation of dried leaves using Clevenger apparatus, British Pharmacopia, Vol. 11.1988 (Appendix XI E A 137E volatile oil in Drug)</td>
</tr>
<tr>
<td>2</td>
<td>Odour</td>
<td>Characteristic</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Oil Percentage</td>
<td>1.4%</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Refractive Index</td>
<td>1.471 at 26.8°C</td>
<td>ISO 280:1999 (E)</td>
</tr>
<tr>
<td>7</td>
<td>Acid Value</td>
<td>1.873</td>
<td>ISO 1242:1999 (E)</td>
</tr>
<tr>
<td>8</td>
<td>Ester Value</td>
<td>17.70</td>
<td>British Standard methods of tests for essential oils (1953)</td>
</tr>
</tbody>
</table>
Thin Layer Chromatography (TLC)

TLC was carried out in the solvent system of n-hexane:ethyl acetate (96:4). Anisaldehyde methanolic sulfuric acid (0.5ml Anisaldehyde + 10 ml Glacial acetic acid + 85 ml Methanol+ 5 ml conc. Sulfuric acid) was used as developing reagent and TLC plate was heated in oven at 110°C for 10 minutes. Four spots were observed with RF values of 0.17, 0.46, 0.61, and 0.84.

GC/MS Analysis

The oil obtained from hydrodistillation was injected on a GCMS – QP 2010 Plus (SHIMADZU), Gas Chromatography Mass Spectrometer instrument using a Rtx-5MS column (30m X 0.25mm X 0.25 μm) and with library NIST 11 and FFNSC 1.3. Carrier gas was Helium.

Results and Discussion

The oil was obtained by conventional hydro distillation of fresh leaves of Citronella in a Clevenger apparatus. The oil percentage obtained was 1.4% on fresh weight basis. During GC-MS analysis, compounds were identified by using MS-library: Nist-11 and FFNSC 1.3. GC-MS analysis of Citronella oil resulted in the identification of following compounds: Nerol (52.97%), Neral (27.43%), Nerol acetate (6.34%) and Cetronellol (3.80%), Cetronellal (2.57%), Cyclohexene, 1-methyl-4-(1-methylethenyl)-(s) (1.82%), Linalool (0.62%), Citronellyl acetate (0.74%), trans-z, alpha-Bisabolene epoxide (0.60%), Isogeranial (0.55%), Hept-5-en-2-oneA6 methylA (0.53%), Alpha-Cadinol (0.40%).

Fig. 1: Showing the temp. program, chromatogram and chemical constituents of citronella oil

<table>
<thead>
<tr>
<th>Peak#</th>
<th>R.Time</th>
<th>Area</th>
<th>Ret. %</th>
<th>Name</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.739</td>
<td>15057</td>
<td>0.53</td>
<td>Hept-5-en-2-one A6 methylA</td>
</tr>
<tr>
<td>2</td>
<td>10.121</td>
<td>533427</td>
<td>1.82</td>
<td>Cyclohexene, 1-methyl-4-(1-methylethenyl)</td>
</tr>
<tr>
<td>3</td>
<td>12.515</td>
<td>299791</td>
<td>1.62</td>
<td>Linalool</td>
</tr>
<tr>
<td>4</td>
<td>14.389</td>
<td>751741</td>
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</tr>
<tr>
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<td>15.401</td>
<td>160667</td>
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<tr>
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<td>Nerol</td>
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<td>18.207</td>
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<td>Nerol</td>
</tr>
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<td>9</td>
<td>18.618</td>
<td>4708703</td>
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<td>2,6-Octadienal, 3,7-dimethyl, (Z)</td>
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<tr>
<td>10</td>
<td>21.139</td>
<td>215530</td>
<td>0.74</td>
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<td>11</td>
<td>22.393</td>
<td>1836922</td>
<td>0.34</td>
<td>2,6-Octadienal (E), 3,7-dimethyl, acetate</td>
</tr>
<tr>
<td>12</td>
<td>23.393</td>
<td>667772</td>
<td>2.17</td>
<td>Biocycl[7.2.0]Heptac-4-ene, 4,11,11-trimethyl-8-methylene (IR:1R&quot;,12R&quot;)</td>
</tr>
<tr>
<td>13</td>
<td>24.236</td>
<td>101238</td>
<td>0.35</td>
<td>Humulene</td>
</tr>
<tr>
<td>14</td>
<td>26.716</td>
<td>176240</td>
<td>0.68</td>
<td>trans-Z, alpha-Dihydrodiene epoxide</td>
</tr>
<tr>
<td>15</td>
<td>27.724</td>
<td>115082</td>
<td>0.40</td>
<td>Alpha-Cadinol</td>
</tr>
</tbody>
</table>

Fig. 1: Showing the temp. program, chromatogram and chemical constituents of citronella oil
Physico – chemical parameters (Specific Gravity: 0.9453 at 24.6 p C, Refractive Index: 1.471 at 26.8 p C, Optical Rotation: -1.16p at 28.2 p C, Acid Value: 1.873 and Ester Value: 17.70) were determined.

**Conclusion**

We conclude that the essential oil of citronella leaves can be isolated by simple hydrodistillation method and thus the chemical composition and other physiochemical parameters were studied. The findings of this study indicate the presence of the major chemical constituents as Nerol, Neral, Nerol acetate and Cetronellol, Cetronellal. The TLC performed of the oil can be henceforth used as reference TLC for further analysis.

**Acknowledgements**

The authors are grateful to the Director General of DPR Mr. Rajdev Prasad yadav, Deputy Director General Mrs. Susma Upadhyaya and Mrs. Jyoti Joshi Bhatta, Chief of NPRL for their great motivation and Mr. Rajendra Sharma, Mr. Krishna Kumar Shah, Mr. Rajeswar Ranjitkar and Mr. Govinda Prasad Gautam are also thankful for their cooperation and encouragement.

**References**


Antifungal activity of Lantana camara L. leaves extract against plant pathogenic fungi

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Abstract

In-vitro studies were carried out to determine the antifungal activity of methanolic extract of leaves of Lantana camara L. against Bipolaris sorokiniana (Sacc.) Shoemaker, Colletotrichum gloeosporioides (Penz.) Penz. & Sacc., Fusarium oxysporum Schltdl., Rhizoctonia solani J.G. Kühn and Verticillium alboatrum Reinke and Berthold by poisoned food technique. The results revealed that methanolic extract of leaves of L. camara had strong antifungal activity with significant inhibition on the growth of B. sorokiniana and V. alboatrum.

Keywords: Lantana camara extract, antifungal activity, poisoned food technique

Introduction

Application of synthetic fungicides has been the primary means for management of crop diseases. However, apart from their high costs, their residues pose potential health hazards and environmental contamination (Okinbo and Osuinde, 2003). Besides, frequent application of fungicides could lead to the development of resistance in pathogen populations (Kumar et al., 2007). Thus, substantial use of chemical pesticides induces problems of health and environmental hazards in agricultural system. Therefore, it is important to find a practical, cost effective and non-toxic method to manage plant diseases. So, for human and plants, natural products of antimicrobial activity are best biorational alternatives today (Tiwari et al., 2007).

Biologically active substances have been reported to be present in various green plants. Plant derived substances have recently become of great interest owing to their versatile applications (Baris et al., 2006). The substances of plant origin have very high potential as pesticides. These substances can not only be easily procured from profusely available plants, but also have an additional advantage of being biodegradable and non-pollutant. Active constituents of the medicinal and aromatic plants have been found to be less phytotoxic, more systemic and easily biodegradable (Fawcett and Spencer, 1970).

Lantana camara L. is a perennial shrub belonging to family Verbenaceae. Due to its resilient nature, the plant is widely distributed in the pantropic. It is a problem weed dominating native species and disrupting biodiversity (Priyansha and Joshi, 2013). It is among top ten invasive weeds on the earth (Sharma et al., 2005). Hence, strategies should be developed to optimize the usefulness of this plant. It has several uses, mainly as herbal medicine and in some areas as firewood and mulch (Sharma et al., 1988; Sharma et al., 1999; Day et al., 2003). This plant, besides being easily available, possesses non-phytotoxic compounds that are found to exhibit inhibitory effect on pathogens (Vaidya and Bhattarai, 2009). It has also shown strong insecticidal and antimicrobial activity in numerous experiments. Storing potatoes with lantana leaves nearly eliminates the damage caused by Phthorimaea operculella Zeller, the potato tuber moth (Lal, 1987). It has been proven to be rich in verbascosides which have strong antifungal properties (Oyourou et al., 2013). Lantana fixed extracts and essential oils have been shown to be effective against plant pathogens and storage fungi such as Alternaria spp. (Srivastava and Singh, 2011), wood rot fungi (Tripathi et al., 2009), Penicillium digitatum (Pers.) Sacc. (Oyourou et al., 2013), Mucor spp. and Fusarium solani (Mart.) Sacc. (Rizvi et al., 2013), Aspergillus niger Tiegh., Penicillium funiculosum Thom, Rhizomucor...
*auricus* and *Trichoderma reesi* (Dharmagadda et al., 2005) etc.

There is a large demand for new fungicides for use in food protection, agriculture and medicine. In recent years there has been a growing trend to evaluate the antimicrobial activity of the extracts and isolates of medicinal plants, because of resistance developed by pathogens, gross side effects of synthetic drugs due to indiscriminate use and their expensive treatment regimen (Nychas, 1995; Tauxe, 1997; Cowan, 1999; Smid and Gorris, 1999; Sheriff, 2001; Tomoko et al., 2002). Hence, this research was conducted to identify environmentally feasible alternatives to synthetic fungicides for crop protection.

**Material and Methods**

**Collection of plant materials**

*L. camara* plants were collected from Sankhamul, Lalitpur in the month of May 2015. After collection, leaves were separated, washed with water and shade dried for 15 days. Then they were powdered in a grinder.

**Preparation of methanolic extract**

The 50 g of powdered leaves were macerated with 300 ml of 80% methanol in a dropping funnel for three days. The extract was collected and the residue was macerated again with 50 ml of 80% methanol for 24 hours. The process was repeated twice. The liquid extract was air dried for one month till thick consistency was obtained.

**Fungal isolates**

Antifungal activity of the *L. camara* extract was evaluated against five species of fungal plant pathogens obtained from Biological Section, Department of Plant Resources. These pathogens had been isolated from different parts of *Swertia chirayita* (Roxb. ex Fleming) Karsten exhibiting various disease symptoms (Table 1).

**Evaluation of antifungal activity by poisoned food technique**

Antifungal activity of the *L. camara* extract was determined by poisoned food technique (Das et al., 2010). Sabouraud Dextrose Agar (SDA) poisoned with *Lantana* extract of 5% strength was prepared by dissolving 5 parts extract to 95 parts of SDA followed by thorough stirring for uniform dissolution. Control medium was prepared by adding distilled water instead of *L. camara* extract in the same ratio. The media were then sterilized at 121°C at 15 psi pressure for 15 minutes. SDA plates were prepared by pouring 15 ml of the sterilized medium into each petri-plate. With a sterile 6 mm cork borer, discs were cut from the growing edge of seven-day-old cultures of each of the five fungal species and each disc was placed at the center of an SDA plate. Each treatment was done in triplicate. The inoculated plates were incubated at 25±2°C for seven days. The diameters of the fungal colonies (linear growth) were measured on second, fifth and seventh day.

The fungitoxicity of the extract in terms of percentage inhibition of mycelial growth was calculated using the following formula:

\[
\text{Percent inhibition} = \frac{C - T}{C} \times 100
\]

where,

\[C = \text{Average diameter of mycelial growth in control plate}\]

\[T = \text{Average diameter of mycelial growth in treatment plate}\]

**Table 1:** Fungal species used for the screening of antifungal activity

<table>
<thead>
<tr>
<th>S.no.</th>
<th>Fungal species</th>
<th>Isolate designation</th>
<th>Source part of <em>S. chirayita</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Bipolaris sorokiniana</em> (Sacc.) Shoemaker</td>
<td>Sc-Ila1-8-Le</td>
<td>Leaf</td>
</tr>
<tr>
<td>2</td>
<td><em>Colletotrichum gloeosporioides</em> (Penz.) Penz. &amp; Sacc.</td>
<td>Sc-Ila2-11-Le</td>
<td>Leaf</td>
</tr>
<tr>
<td>3</td>
<td><em>Fusarium oxysporum</em> Schldl.</td>
<td>Sc-Ila1-1-Ro</td>
<td>Root</td>
</tr>
<tr>
<td>4</td>
<td><em>Rhizoctonia solani</em> J.G. Kühn</td>
<td>Sc-Ila1-5-St</td>
<td>Stem</td>
</tr>
<tr>
<td>5</td>
<td><em>Verticillium albovatum</em> Reinke &amp; Berthold</td>
<td>Sc-Ila1-1-Ro</td>
<td>Root</td>
</tr>
</tbody>
</table>
Statistical analysis
All the tests were conducted in triplicates and observations were expressed as mean ± standard error (SE). The linear growths (colony diameters) of the test fungi in poisoned medium and control condition were compared by paired-samples t test. The percent inhibitions of the fungal growths of the test fungi by the L. camara extract were compared by Tukey’s HSD test.

Result and Discussion
In the present study, antifungal activity of methanol extract of L. camara against five different fungal species was studied by poisoned food technique (Table 2 and 3, Figure 1).

The comparison of the colony diameter of different test fungi grown in L. camara-extract-poisoned medium and in control condition showed that the linear growths of B. sorokiniana, F. oxysporum and V. alboatrum in poisoned medium was significantly different in comparison to their growths in control condition on second, fifth as well as seventh days. However, linear growth of C. gloeosporoides was not affected significantly. In case of R. solani, the growth on treated and control conditions differed significantly on second day. The fifth and seventh day observations for R. solani could not be compared since the diameter of colonies formed under both treated and control conditions had exceeded the petridish diameter (9 cm) (Table 2).

On comparing percentage inhibition of the five test species of fungi by L. camara extract, it was found that, on second day, R. solani growth was best inhibited followed by B. sorokiniana and V. alboatrum, while F. solani was least inhibited. Similarly on fifth day, B. sorokiniana was found to be most inhibited followed by V. alboatrum and F. oxysporum. Similar trend was also observed on seventh day (Plates 1a to 1e). However, negative inhibition was observed in case of C. gloeosporoides on second and fifth day i.e. its growth was promoted by L. camara extract (Table 3 and Figure 1).

Fungitoxic effects of fixed extracts of L. camara has been reported against several fungal plant pathogens. Naz and Bano (2013) evaluated antimicrobial potential of extracts of L. camara in different

Table 2: Comparison of colony growth fungi in poisoned media and control condition

<table>
<thead>
<tr>
<th>S.no.</th>
<th>Fungi</th>
<th>Mean colony diameter±SE (cm)</th>
<th>Sig. (2-tailed)*</th>
<th>Difference at 5% level of significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Poisoned medium</td>
<td>Control</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Bipolaris sorokiniana (Sacc.)</td>
<td>1.85 ± .03</td>
<td>2.60 ± .03</td>
<td>.006</td>
</tr>
<tr>
<td>2</td>
<td>B. sorokiniana on day 5</td>
<td>3.15 ± .03</td>
<td>5.95 ± .03</td>
<td>.000</td>
</tr>
<tr>
<td>3</td>
<td>B. sorokiniana on day 7</td>
<td>4.00 ± .05</td>
<td>7.85 ± .03</td>
<td>.000</td>
</tr>
<tr>
<td>4</td>
<td>Colletotrichum gloeosporiodes (Penz.)</td>
<td>1.63 ± .07</td>
<td>1.48 ± .03</td>
<td>.188</td>
</tr>
<tr>
<td>5</td>
<td>C. gloeosporioides on day 5</td>
<td>3.37 ± .07</td>
<td>3.25 ± .03</td>
<td>.250</td>
</tr>
<tr>
<td>6</td>
<td>C. gloeosporioides on day 7</td>
<td>4.60 ± .05</td>
<td>4.65 ± .03</td>
<td>.478</td>
</tr>
<tr>
<td>7</td>
<td>Fusarium oxysporum Schltld. on day 2</td>
<td>1.70 ± .05</td>
<td>2.02 ± .02</td>
<td>.019</td>
</tr>
<tr>
<td>8</td>
<td>F. oxysporum on day 5</td>
<td>4.83 ± .04</td>
<td>5.55 ± .06</td>
<td>.004</td>
</tr>
<tr>
<td>9</td>
<td>F. oxysporum on day 7</td>
<td>6.93 ± .02</td>
<td>7.75 ± .03</td>
<td>.002</td>
</tr>
<tr>
<td>10</td>
<td>Rhizoctonia solani J.G. Kühn on day 2</td>
<td>2.78 ± .38</td>
<td>7.47 ± .02</td>
<td>.006</td>
</tr>
<tr>
<td>11</td>
<td>R. solani on day 5</td>
<td>9.00 ± .00</td>
<td>9.00 ± .00</td>
<td>could not be interpreted</td>
</tr>
<tr>
<td>12</td>
<td>R. solani on day 7</td>
<td>9.00 ± .00</td>
<td>9.00 ± .00</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>Verticillium alboatrum Reinke &amp; Berthold on Day 2</td>
<td>1.55 ± .10</td>
<td>2.12 ± .02</td>
<td>.021</td>
</tr>
<tr>
<td>14</td>
<td>V. alboatrum on Day 5</td>
<td>4.52 ± .03</td>
<td>5.57 ± .02</td>
<td>.002</td>
</tr>
<tr>
<td>15</td>
<td>V. alboatrum on Day 7</td>
<td>6.32 ± .07</td>
<td>7.58 ± .04</td>
<td>.007</td>
</tr>
</tbody>
</table>

* sig. value generated by paired sample T test
solvents and concluded that the methanolic leaf extract exhibited significant inhibition against *Aspergillus flavus* Link and *Aspergillus fumigatus* Fresen. Srivastava and Singh (2011) have reported significant antifungal activity of *L. camara* extracts against *Alternaria* sp. Tripathi et al. (2009) demonstrated efficient antifungal activity of ethanol and hot water extract of this plant against wood destroying white and brown rot fungi. Sharma and Kumar (2009) have suggested that *L. camara* extracts can be used to manage *Fusarium oxysporum* Schltdl., an important destructive soil borne pathogen. Sailaja (2014) has reported prominent antifungal activity of *L. camara* extracts against *Aspergillus niger* Tiegh. and *Candida albicans* (C.P. Robin) Berkhout.

**Conclusion**

The results obtained from this study indicate that methanolic extract of *L. camara* has strong antifungal activity against two fungal pathogens viz. *B. sorokiniana* and *V. alboatrum*. The extract also showed moderate antifungal activity against *F. oxysporum*. Hence, it can be concluded that highly effective ecofriendly fungicidal agents can be developed by optimizing the extraction and separation procedure of *L. camara* extracts.

**Acknowledgment**

We would like to express our sincere gratitude to Mr. Rajdev Prasad Yadav, Director General, Department of Plant Resources for providing us an excellent opportunity to conduct this research. We are also deeply indebted to Ms. Sushma Upadhyay and Mr. Sanjeev Kumar Rai, Deputy Director Generals, Department of Plant Resources for encouraging us in our research activities.

**References**


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**Table 3:** Percent of inhibition shown by leaves of 80% methanolic extract of *Lantana camara* L. against five different plant pathogenic fungi

| S.no. | Fungus species               | Percentage inhibition ± SE*  
|-------|-----------------------------|-------------------------------
|       |                             | on 2nd day | on 5th day | on 7th day |
| 1     | *Bipolaris sorokiniana* (Sacc.) Shoemaker | 28.85 ± 1.11* | 47.06 ± 0.49* | 49.04 ± 0.64* |
| 2     | *Colletotrichum gloeosporioides* (Penz.) Penz. & Sacc. | -10.11 ± 4.90* | -3.59 ± 2.05* | 1.08 ± 1.08* |
| 3     | *Fusarium oxysporum* Schltdl. | 15.70 ± 2.48* | 12.91 ± 0.79* | 10.54 ± 0.22* |
| 4     | *Rhizoctonia solani* J.G. Kühn | 62.72 ± 5.13* |            |             |
| 5     | *Verticillium alboatrum* Reinke & Berthold | 26.77 ± 4.72* | 18.86 ± 0.60* | 16.70 ± 0.88* |
| 6     | Control treatment            | 0.00 ± 0.00* | 0.00 ± 0.00* | 0.00 ± 0.00* |

*Grouping on the basis of Tukey’s HSD at 5% level of significance. In columns, the means followed by the same letters are not significantly different (P=0.05).


Plate 1: Growth of test fungi in *L. camara*-extract-treated and control medium

- **a. Bipolaris sorokiniana**
- **b. Colletotrichum gloeosporioides**
- **c. Fusarium oxysporum**
- **d. Rhizoctonia solani**
- **e. Verticillium alboatrum**
Acclimatization of two epiphytic orchids: *Coelogyne stricta* (D. Don) Schltr. and *Coelogyne flaccida* Lindl. propagated under *in vitro* conditions

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² Plant Biotechnology and Biochemistry Unit, Central Department of Botany, Tribhuvan University, Kirtipur. 
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Abstract

*In vitro* propagation is the best alternative method for conservation of rare and endangered orchids. Nevertheless, its wider implication is restricted due to high percentage of plant loss during transferring to *ex vitro* condition. Hence, the goal of this study was to find out the best acclimatization substrates for *Coelogyne stricta* (D. Don) Schltr. and *Coelogyne flaccida* Lindl. grown under *in vitro* conditions. The acclimatization procedure included gradual hardening from incubation room to natural environment. *In vitro* grown plantlets having well developed shoots & roots and measuring more than 2.5 cm in height were subsequently transferred to different acclimatization substrates/potting mixture for acclimatization. Plantlets of *C. stricta* showed best survival rate of 90% in potting mixture consisting of coco-peat + sphagnum moss + sand in the ratio of 2:1:1 followed by 80% in tree fern powder + sand in the ratio of 2:1. Similarly, *C. flaccida* showed best survival rate of 80% in the potting mixture consisting of tree fern powder + sphagnum moss in the ratio of 2:1 followed by 70% in coco-peat + sphagnum moss + sand in the ratio of 2:1:1.

Keywords: Acclimatization, *in vitro*, potting mixture, *Coelogyne stricta*, *Coelogyne flaccida*.

Introduction

*In vitro* propagation has been extensively utilized for rapid and mass multiplication of many plant species including orchids. During the *in vitro* stage, plants are kept in a controlled environment, characterized by high humidity, low light intensity, supplementary sugar supply and growth regulators (Murashige 1974; Viegas et al. 2005). However, its application is restricted often by the high percentage rate of plant loss when transferred to *ex vitro* condition. This is due to plantlets need to get adjusted with many harsh conditions of *ex vitro* environment like high level of irradiance, low humidity and limiting water due to low hydraulic conductivity of roots and root-stem connections (Fila et al. 1998). Acclimatization of the plantlets can overcome these problems with the subsequent use of green house and gradual lowering in air humidity (Lavanya et al. 2009). The development of successful hardening technique is prerequisite for *in vitro* propagation method. Hardening accounts for about 60% of the total production cost (Hazarika 2003). Therefore, an efficient and cost-effective acclimatization technique is necessary for *in vitro* raised plantlets. So, we endeavored to develop a new efficient and cost-effective hardening technique for *in vitro* raised plantlets of *Coelogyne stricta* (D. Don) Schltr. and *Coelogyne flaccida* Lindl. They both are ornamental and medicinal orchids belonging to family Orchidaceae.

*Coelogyne stricta*, a native orchid of Nepal, is commonly known as ‘The Rigid Coelogyne Pseudobulb’ and is an epiphyte found on tree trunks or lithophytes on mossy rocks at elevations of 1400-2000 m (Raskoti 2009; Rajbhandari 2015). It has high aesthetic value (Figure 1) so it is used as an ornamental plant in different gardens, nurseries, hotels, etc. Its medicinal value is due to paste of its pseudobulb which is applied to the forehead against headache and fever (Baral and Kurmi 2006; Pant and Raskoti 2013).
Coelogyne flaccida, a native orchid of Nepal, is commonly known as ‘The Loose Coelogyne Pseudobulb’ and is an epiphyte found on tree trunks at elevations of 900-1100 m (Raskoti 2009; Rajbhandari 2015). It also has high aesthetic value (Figure 2) so is also used as an ornamental plant in different gardens, nurseries, hotels, etc. Its medicinal value resides on the paste of its pseudobulb, which is used for headache treatment; and the juice is convenient for indigestion relief (Manandhar 2002; Pant and Raskoti 2013).

Both of these orchid species are vanishing rapidly from their natural habitat and are restricted to narrow areas due to their uncontrolled collection, illegal trade, deforestation, habitat destruction and high price in the national and international markets. So, various tissue culture protocols have been developed for large scale production of both these orchids using their seeds and pseudo-bulbs as explants.

Nevertheless, no acclimatization techniques for these orchids have been developed for in vitro propagated plantlets. So, from both conservation and commercial point of view it is imperative to develop acclimatization techniques of these orchids using standard method. Therefore, the present study was undertaken to optimize the acclimatization of in vitro raised C. stricta and C. flaccida as the final stage of successful in vitro propagation. Eight different potting mixture were tested in this study.

Materials and methods

The in vitro grown plantlets of Coelogyne stricta (D. Don) Schltr. (Figure 3) and Coelogyne flaccida Lindl. (Figure 4) having well developed shoots and roots and measuring more than 2.5 cm in height maintained at 25± 2ÚC and 16 hours photoperiod were taken from incubation room of Tissue Culture Section of National Herbarium and Plant Laboratories, Godawari, Lalitpur.
The jars containing well developed plantlets were opened and kept at room temperature for one week. The plantlets were then taken out from the jars and washed in running tap water thoroughly to remove traces of adhered medium and agar from the surface of plantlets. The plantlets were rinsed in bavistin (0.1%, w/v) for five minutes to kill fungus if present and to minimize the chances of infection. After that the plantlets were washed with distilled water and dried using blotting paper.

The plantlets were transferred to cleaned plastic trays containing different potting mixtures (Table 1 & 2). Ten plantlets of both orchids were tested against each combination of potting mixture separately. Coco-peat and tree fern powder used for the potting mixture were made wet using tap water prior to use. The sand used was washed with water and made dry under the direct exposure of sunlight.

NPK solution (5%) was sprayed once a week for fastening their growth. The plants were then covered using transparent polythene sheets, with holes made for aeration, to control humidity. The plants were kept in greenhouse for three months and finally transferred to the natural environment.

NPK solution (5%) was sprayed once a week for fastening their growth. The plants were then covered using transparent polythene sheets, with holes made for aeration, to control humidity. The plants were kept in greenhouse for three months and finally transferred to the natural environment.

**Results and Discussion**

The well developed *in vitro* grown plantlets of *Coelogyne stricta* (D. Don) Schltr. and *Coelogyne flaccida* Lindl. measuring more than 2.5 cm in height showed best survival rate of 90% and 80% on potting mixture consisting of coco-peat + sphagnum moss + sand (2:1:1) and tree fern powder + sphagnum moss (2:1) respectively (Table 1 & 2). It suggests the effectiveness of coco-peat, tree fern powder and sphagnum moss in the successful acclimatization of *in vitro* grown epiphytic orchids. Coco-peat, tree fern powder and sphagnum moss were used because of their high water holding capacity to provide moisture to the plantlets and sand might provide the heat needed for the initiation of new roots that is suitable to adapt to the natural environment.

Franco et al. (2007) in the acclimatization of *Cattleya trianae* found substrate of pine-coconut (1:1), coconut fiber alone and pine-coconut-coal (1:1:1) had the percent survival of 80, 76 and 60% respectively while the lowest percentage survival was recorded for pine bark at 12%. Similarly, Das et al. (2007) found 90% of *Cymbidium devonianum* plantlets survived on substratum containing brick, charcoal and decaying litter in the ratio of (1:1:1) with a layer of moss on top while Dutra et al. (2009) found 90% survival of the 35 weeks old seedlings of *Cyrtopodium punctatum* when potted in coconut hush rowing medium.

### Table 1: Acclimatization of *Coelogyne stricta* (D. Don) Schltr. in different potting mixture

<table>
<thead>
<tr>
<th>Potting mixture</th>
<th>Ratio of potting mixture</th>
<th>Total number of plantlets</th>
<th>No. of plantlets hardened after</th>
<th>% of acclimatization</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>2nd week</td>
<td>4th week</td>
</tr>
<tr>
<td>Coco-peat + sphagnum moss</td>
<td>2:1</td>
<td>10</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>Coco-peat + sphagnum moss + sand</td>
<td>2:1:1</td>
<td>10</td>
<td>10</td>
<td>9</td>
</tr>
<tr>
<td>Coco-peat + sphagnum moss + sand + soil</td>
<td>2:1:1:1</td>
<td>10</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Coco-peat + sand</td>
<td>2:1</td>
<td>10</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Coco-peat + sand + soil</td>
<td>2:1:1</td>
<td>10</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Tree fern powder + sphagnum moss</td>
<td>2:1</td>
<td>10</td>
<td>8</td>
<td>6</td>
</tr>
<tr>
<td>Tree fern powder + sphagnum moss + sand</td>
<td>2:1:1</td>
<td>10</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Tree fern powder + sand</td>
<td>2:1</td>
<td>10</td>
<td>10</td>
<td>8</td>
</tr>
</tbody>
</table>
Kaur and Bhutani (2009) recorded nearly 75% of plantlets survival of *Vanda testacea* when transferred to pots containing epiphytic compost (1 charcoal: 1 brick pieces: 1 bats). Pant and Shrestha (2011) successfully hardened nearly 70% *Phaius tancarvilleae* on potting mixture containing coco-peat and sphagnum moss (2:1). Gogoi et al. (2012) in *Cymbidium eburneum* reported survivability of 70% when complete plantlets were grown in the compost mixture comprising brick, charcoal and decaying litter in the ratio 1:1:1 and a layer of moss on top.

Pant and Thapa (2012) in *Dendrobium primulinum* successfully hardened nearly 70% in the potting mixture containing coco-peat and sphagnum moss in the ratio of 2:1 while Paudel and Pant (2012) in *Esmeralda clarkei* reported 85% survival in potting mixture consisting of soil, sand and sawdust (1:1:1). Pradhan et al. (2013) in *Dendrobium densiflorum* found out 85% survival in coco-peat, litter and clay (2:1:1) while Warghat et al. (2014) found cent percent survival in *Dactylorhiza hatagirea* when grown in potting mixture consisting of coco-peat + vermiculite + perlite (1:1:1).

Table 2: Acclimatization of *Coelogynestricta* (D. Don) Schltr. and *Coelogyne flaccida* Lindl. in different potting mixture

<table>
<thead>
<tr>
<th>Potting mixture</th>
<th>Ratio of potting mixture</th>
<th>Total number of plantlets</th>
<th>2nd week</th>
<th>4th week</th>
<th>6th week</th>
<th>8th week</th>
<th>% of acclimatization</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coco-peat + sphagnum moss</td>
<td>2:1</td>
<td>10</td>
<td>7</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>60</td>
</tr>
<tr>
<td>Coco-peat + sphagnum moss + sand</td>
<td>2:1:1</td>
<td>10</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>70</td>
</tr>
<tr>
<td>Coco-peat + sphagnum moss + sand + soil</td>
<td>2:1:1:1</td>
<td>10</td>
<td>6</td>
<td>6</td>
<td>5</td>
<td>5</td>
<td>50</td>
</tr>
<tr>
<td>Coco-peat + sand</td>
<td>2:1</td>
<td>10</td>
<td>7</td>
<td>6</td>
<td>6</td>
<td>5</td>
<td>50</td>
</tr>
<tr>
<td>Coco-peat + sand + soil</td>
<td>2:1:1</td>
<td>10</td>
<td>6</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>40</td>
</tr>
<tr>
<td>Tree fern powder + sphagnum moss</td>
<td>2:1</td>
<td>10</td>
<td>8</td>
<td>8</td>
<td>8</td>
<td>8</td>
<td>80</td>
</tr>
<tr>
<td>Tree fern powder + sphagnum moss + sand</td>
<td>2:1:1</td>
<td>10</td>
<td>7</td>
<td>7</td>
<td>6</td>
<td>6</td>
<td>60</td>
</tr>
<tr>
<td>Tree fern powder + sand</td>
<td>2:1</td>
<td>10</td>
<td>7</td>
<td>6</td>
<td>5</td>
<td>5</td>
<td>50</td>
</tr>
</tbody>
</table>

Figure 5: Comparative percentage acclimatization of *Coelogyne stricta* and *Coelogyne flaccida* in different potting mixture.
Optimum acclimatization substrate/potting mixture may vary for different species, and so for intra-generic species. Different optimum potting mixture of these orchid species might be due to their different genetic constitution, physiological state and endogenous growth regulators present in them. After the hardening phase, the plants were taken to a greenhouse (Figure 6 & 7) and later to the natural environment on their host trees where they showed qualitative characteristics such as vigor, hardness and waxy texture, green coloration in the leaves and velamen formation.

**Acknowledgements**

The authors would like to express sincere gratitude to Dr. Sushim Ranjan Baral, former Chief, National Herbarium and Plant Laboratories, Godawari, Lalitpur and Prof. Dr. Pramod K. Jha, Head, Central Department of Botany, Tribhuvan University, Kirtipur for arranging and providing necessary laboratory facilities for this research.

**Reference**


**Conclusions**

The best acclimatization substrate for plantlets of *Coelogyne stricta* was found to be potting mixture consisting of coco-peat + sphagnum moss + sand in the ratio of 2:1:1 with survival rate of 90% while *C. flaccida* showed best survival rate of 80% in the potting mixture consisting of tree fern powder + sphagnum moss in the ratio of 2:1.


Micropropagation of **Pogostemon cablin** (Benth.) (Patchouli)

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Abstract

The present study aims to investigate an efficient protocol of micropropagation of **Pogostemon cablin** (Benth.). It is commercially important plant for its aromatic patchouli oil. It was newly introduced in Nepal from Assam, India by Chaudhary Biosys Pvt. Ltd, Nepal. Plants were regenerated using leaf and shoot tip explants in Murashige and Skoog (MS) medium supplemented with phyto hormones BAP and NAA and the medium was also fortified with 3% sucrose and 0.8% agar. The best multiplication was found in lower concentration of BAP (i.e. 0.1 to 1mg/l ) with 0.1ml/l NAA. For rooting, the in vitro multiplied micro shoots were acclimatized and transferred to non sterile sands for induction of roots maintaining humidity in poly house. After four weeks, the microshoots induced roots in sand. The rooted plants were grown in polybags filled with soil for further growth.

**Key words:** **Pogostemon cablin**, explants, Benzyl amino purine, Naphthalene acetic acid, shoot proliferation, micro propagation

Introduction

Patchouli (**Pogostemon cablin** Benth.) is commercially important plant for its aromatic patchouli oil. Patchouli belongs to family Lamiaceae. Its oil is commercially used in perfumes and cosmetics (Hasegawa *et al*., 1992; Maheswari *et al*., 1993). It also possess ant insecticidal, antifungal and bacteriostatic properties (Kukreja *et al*., 1990, Yang 1996, Pattnaik *et al*. 1996). In aromatherapy it is used to calm nerve, relief depression and stress. The oil extensively used as a flavoring ingredient in major food products, including alcoholic and non-alcoholic beverages, frozen dairy desserts, candy, baked goods, gelatin, meat and meat products. The oil is regarded as the best fixative for heavy perfumes which imparts strength, character, alluring notes and lasting qualities, (Faroqui and Sreeramu 2001). Because of shade loving crop it can be grown as an intercrop with other trees. It is native plant of tropical area and widely grown to India, Maleysia, Indonesis Philippines and Singapore.

Conventionally, the vegetative propagation was done by stem cuttings and also from seeds. Propagation from seeds could not produce true to type plants and propagation from stem cutting could not produce sufficient amount of plants. Its need more space and time. In Nepal, it is newly introduced plants from Assam, India by Chaudhary Biosys Pvt. Ltd., Nepal (Chaudhary Biosys Pvt. Ltd Nepal is a plant oil exporters in Nepal. It was established in 2004 to collectively work with INGO’s, Government Agencies and Communities in the field of Non-Timber Forest Product (NTFP) by establishing a world class Medicinal and Aromatic Plant based Processing Industry to uplift the less privileged community in Nepal).It was cultivated in Kakervitta, Jhapa district and in Chitwan district . The distillation unit was also established in Jhapa district. Many
farmers have shown their interest to cultivate this plant. Tissue culture method is an alternative method for large scale commercial production. Hence the present study aims to develop an efficient method of micro propagation using nodal and leaf explants for the production of patchouli plants to mitigate the demands of farmers.

**Material and Method**

The elite plants were brought from Chaudhary Biosys Pvt. Ltd., Nepal and planted in pots as mother stocks. The shoot tips were taken from these plants, washed in running tap water for one hour with few drops of teepol and again washed with distilled water. These shoot tips were surface sterilized with 0.1% mercuric chloride solution for 5 minutes and washed with sterilized distilled water for five times. Aseptically, unnecessary parts from these explants were cut down by sharp sterilized blade into small shoot tips (1-2mm) and also into single leaves as explants. These shoot tips and single leaves were then cultured in MS medium with different concentration of hormones. Ten replicates were made in each concentration of BAP and NAA. The medium was also fortified with 0.1% Casein hydrolysate, 3% sucrose and 8% agar for solidification of medium. The pH of the medium was adjusted to 5.5 before autoclaving. The cultures were then incubated at 25±2 °C under 16 hour photoperiod under 3 kilo lux light intensity provided by white florescence tubes.

The proliferated micro shoots from these shoot tips and leaves were again subcultured in MS medium with different concentration of BAP and NAA. Among the different concentration of BAP and NAA, best hormone concentration was identified and standardized. The microshoots proliferated were subcultured in the same medium for large scale propagation.

For rooting of microshoots, the cultured bottles were acclimatized in poly house for a week. Then the microshoots were removed from the culture flasks, washed thoroughly to remove the medium, cut down 2.5 to 3cm long and were transferred on propagator filled with non-sterile sands (Rajbhandary & Bajaj, 1991) maintaining the humidity in poly house. After two weeks, the microshoots regenerated roots. The rooted plantlets were transferred to polybags with soil for further growth.

**Result**

The shoot tip cultured in MS medium with different concentration of hormones showed different response on different hormones concentration after 4 weeks of culture (Table -1). After four weeks of culture, the shoot tip explants began to regenerate microshoots by direct organogenesis in the MS medium containing 0.1 to 1mg/l BAP in combination with 0.1mg/l NAA. The best multiplication was found in the MS medium containing 1mg/l BAP and 0.1mg/l NAA (Fig-1). In higher concentration of hormone (2mg/l BAP with 0.1mg/l NAA), callus was regenerated at the base along with microshoots (Table-1). The leaf explants began to regenerate microshoots by direct organogenesis from the end of the petiole in the medium 1mg/l BAP with 0.1mg/l NAA after five weeks of culture (Fig-2).

The regenerated micro shoots on different concentration of hormones were again subcultured on MS medium different concentration of BAP with 0.1mg/l NAA and medium without hormone (Table 2). In tested four concentration of BAP and NAA,

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Conc. of growth hormone(mg/l)</th>
<th>Average no. of micro shoots</th>
<th>Callus formation</th>
<th>Ht. of micro shoots in cm</th>
<th>Condition of micro shoots</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>BAP</td>
<td>NAA</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>0.1</td>
<td>0.1</td>
<td>10 to12</td>
<td>A bsence of callus</td>
<td>1 to1. 3</td>
</tr>
<tr>
<td>2</td>
<td>0.5</td>
<td>0.1</td>
<td>10 to12</td>
<td>A bsence of callus</td>
<td>1 to1. 5</td>
</tr>
<tr>
<td>3</td>
<td>1</td>
<td>0.1</td>
<td>15 to 18</td>
<td>A bsence of callus</td>
<td>1 to 1. 5</td>
</tr>
<tr>
<td>4</td>
<td>2</td>
<td>0.1</td>
<td>Stunted 6 to 8</td>
<td>Callus at base</td>
<td>1 to 0.5</td>
</tr>
<tr>
<td>5</td>
<td>MS Medium without hormone</td>
<td>3 to 5</td>
<td>A bsence of callus</td>
<td>1 to 1. 5</td>
<td>Weak</td>
</tr>
</tbody>
</table>

**Table 1 :** Shoot tips responses in MS medium with different concentration of BAP along with 0.1mg/l NAA.
the best multiplication of micro shoots was observed in the MS medium containing 0.5 mg/l BAP with 0.1 mg/l NAA (Fig-3).

For rooting, the subcultured bottles were acclimatized in poly house. The micros hoots were taken out from bottle, washed thoroughly with tap water and 2.5 cm to 3cm long shoots were transferred to sand (Fig. 4). The shoots developed roots within four weeks (Fig. 5). The rooted plantlets were then transferred in poly bags for further growth. Then the micropropagated plants were ready for field transfer (Fig. 6).

### Discussions

Micropropagation of Patchouli plant has been developed by many researchers for many purposes. Misra M (1996) used leaf and node explants of Patchouli for shoot multiplication on MS medium with BA at 1.0 mg/L. He found that shoot formation frequency was maximum (83%) with BA at 1.0 mg/L. Similarly, Maruyama et al. (1997) inoculated node explants on MS medium fortified with 2 µM/l BA which initiated multiple shoots after 25 days. In vitro initiated nodes were used for encapsulation. M. Kumaraswamy and M. Anuradha (2010) obtained multiple shoots from nodal explants in MS medium supplemented with 0.5 mg/l BAP. They found that within four weeks of culture, explants regenerated multiple shoots which attained a height of 3.6 cm. Hua Jin et al. (2014) found that nodal stem with a single node (the second or third node of in vitro plantlets) was the most responsive explants in the MS medium with BA at 0.1–0.2 mg/l. They also pointed out that combinations of BA and NAA resulted in slower shoot development and growth as compared to BA alone. But in our experiment, we used BAP along with NAA for the proliferation of microshoots from shoot tip and leaf explant.

For rooting, most of the authors used half strength MS medium with auxin. Hua Jin et al., (2014) reported that half strength MS medium with 0.2 mg/l IBA was effective for rooting. M. Kumaraswamy and M. Anuradha (2010) rooted shoots in half strength MS medium with 100mg/l activated charcoal. In our experiment, we used non sterile sands for rooting maintaining the humidity of propagator in polyhouse.

### Conclusion

The current study provided information about the efficient protocol of micropropagation for commercial production by in vitro method of *Pogostemon cablin*, which fulfilled the demand of newly introduced plant in Nepal.

### Acknowledgement

The authors are grateful to Mr. Rajdev Prasad Yadav, Director General, Department of Plant Resources, Mr. Y. M. Thapa, former Director General, Department of Plant Resources, Mrs Susma Upadhy and Mr. Sanjeev Kumar Rai, Deputy Director Generals, Department of Plant Resources for their encouragement and providing facilities for this work. We would like to thanks to Mr. Puskar Basnet, Miss Suja Maharjan, Miss Babita Pokhare land Miss Meena Karki for helping during this research works.
References


Micropropagation of *Pogostemon cablin* (Benth.) (Patchouli)

**Figure 1:** Proliferation from shoot tip explant after four weeks

**Figure 2:** Leaf explant in MS medium supplemented with 1.0 mg/l BAP +0.1 mg/l NAA

**Figure 3:** Shoot tip explant in MS medium supplemented with 0.5 mg/l BAP +0.1 mg/l NAA

**Figure 4:** Microshoots transferred in non sterile sands for rooting

**Figure 5:** Sand rooted plants

**Figure 6:** Plants in polybags
Clonal propagation of *Gmelina arborea* Roxb. by Nodal culture

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sabari_rajbahak@yahoo.com

Abstract

Primary cultures were established with nodal segments from juvenile shoots of 6 month old nursery grown *Gmelina arborea* Roxb. Plants. These nodal explant were cultured on a MS medium supplemented with BAP (1.0 mg/L) + NAA(0.1 mg/l) and sucrose (3%) produced optimum bud break in nodal explants. 2-4 shoots were produce from single node explant after 4 weeks of inoculation. Adventitious buds, between 6-8 were formed when in vitro derived single node explants were subcultured on same MS medium. Rooting and field establishment work was under research.

Key words: *Gmelina arborea*, Nodal explants, growth hormones, shoot proliferation, micro propagation

Introduction

The species *Gmelina arborea* Roxb. is an economically and also medicinally important forest tree. *Gmelina arborea* Roxb. locally known as gamhar, Khamari was multipurpose tree which belongs to family Lamiaceae. It is found in sub tropical and temperate forest of Nepal up to 1,200 metres above sea level. Gamhar has also been introduced to most tropical countries as a timber tree.

It was a fast-growing deciduous tall tree attains height up to 30 m with girth of 1.2 to 4 high on favourable sites. *Gmelina* grow best on freely drained, fertile soils, with no hardpan or other impediment to root development. (http://www.Forestrynepal.org resources/trees/Gmelina arborea). Flowering takes place during February to April when the tree is more or less leafless whereas fruiting starts from May onwards up to June. Frequently found in *Shorea robusta* forest, especially the wetter types. Outside Nepal it extends from Pakistan to Vietnam and southern China. (http://www.Forestrynepal.org resources/trees/Gmelina arborea). It is widely planted in tropical countries as a fast-growing timber tree, and in particular as a source of wood for paper pulp.

*Gmelina arborea* timber is reasonably strong for its weight. Once seasoned, it is a very steady timber and moderately resistant to decay and ranges from very resistant to moderately resistant to termites. Its timber is highly esteemed for door and window panels, joinery and furniture especially for drawers, wardrobes, cupboards, kitchen and camp furniture, and musical instruments because of its lightweight, stability and durability. *Gmelina arborea* is a popular timber for picture and slate frames, various types of brush backs, brush handles and toys also for handles of chisels, files, saws, screw drivers, sickles etc. It is also used in artificial limbs, carriages and bobbins. Gamhar is used in papermaking and matchwood industry too.

Planting *G. arborea* with crops like maize and cassava has been found beneficial in increasing the simultaneous production of wood and food. When intercropped with maize and cassava, it performs better under closely stocked stands of cassava, yams and maize. Micropropagation technique plays an important role in conservation and multiplication and also large scale production of an forest tree species. This technique is most valuable for true-to-type of plant production. Conventionally this tree can be propagated by seedlings, stem cuttings, and grafting. There are many factors, which could lead to failure of rooting in stem cuttings. Now a days people are interested for commercial plantation of forest tree species but planting material is not available in sufficient amount so that , this research work was carried out for mass propagation to fulfill growing demand of tree plantation.
Material and Method

*Gmelina arborea* plant was brought from Jhapa district and grown in Biotechnology nursery at Thapathali. 1-2 cm long shoots were cut from 6 month old juvenile plants of *Gmelina arborea*. The shoots were surface sterilized first by washing in running tap water for about half an hour with few drops of liquid detergent Tween-20. After washing the explants were thoroughly rinsed with distilled water for 4-5 times. Further sterilization was done inside a laminar air flow. Explants were surface sterilized with freshly prepared 0.1 % w/v aqueous solution of Mercuric chloride for 5-7 minutes. The explants were then thoroughly rinsed for 3-4 times with sterile distilled water.

Single or double nodal explants were inoculated onto MS basal (Murashige and Skoog, 1962) medium supplemented with different combination of plant growth regulators Benzyl amino purine (1.0 mg/l, 1.5mg/l, 2.0 mg/l, 2.5 mg/l and 3.0mg/l) and Naphthalene acetic acid (0.1 mg/l) for bud break and shoot proliferation. Sucrose 3% was used as carbon sources and media were adjusted to pH 5.8 before sterilized. The media were solidified with 0.8% agar and were autoclaved at 121 °C/15 lbsi.

Before inoculation, explants were transferred to sterilize Petri dish with the help of sterile forceps under strict aseptic conditions. The leaves were removed and single node about 0.5 to 1.0 cm long was transferred to culture bottles containing MS medium with different concentration of growth hormone, BAP and NAA (Figure 1). The cultures were incubated under 16 hour photoperiod with light intensity of 3000 lux florescent tube light and temperature of 25± 2°C.

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Table 1: Effect of different concentration of growth hormone level on Gmelina arborea Roxb. shoot multiplication

<table>
<thead>
<tr>
<th>S.No.</th>
<th>BAP + NAA mg/l</th>
<th>After 4 weeks</th>
<th>After 6 weeks</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>No. of Shoot per explant</td>
<td>No. of Node per shoot</td>
<td>No. of Shoot per node</td>
</tr>
<tr>
<td>1</td>
<td>1.0 + 0.1</td>
<td>2-4</td>
<td>2-3</td>
<td>6-8</td>
</tr>
<tr>
<td>2</td>
<td>1.5 + 0.1</td>
<td>2-3</td>
<td>2-3</td>
<td>2-3</td>
</tr>
<tr>
<td>3</td>
<td>2.0 + 0.1</td>
<td>1-2</td>
<td>1-2</td>
<td>2-3</td>
</tr>
<tr>
<td>4</td>
<td>2.5 + 0.1</td>
<td>1-2</td>
<td>2-3</td>
<td>2-3</td>
</tr>
<tr>
<td>5</td>
<td>3.0 + 0.1</td>
<td>No shoot</td>
<td></td>
<td>-</td>
</tr>
</tbody>
</table>

Clonal propagation of *Gmelina arborea* Roxb. by Nodal culture

Result

Shoot proliferation

After four weeks of inoculation, explants started to show signs of shoot initiation. 2-4 new shoots were produced from the nodal explant. (Figure 2) The micro shoots with 3-4 node were subculture onto the MS medium with same concentration of growth. After 6 weeks 6-8 shoots were regenerate from single node. (Figure 3) The culture was maintained by regular sub culturing at 4 week of intervals to fresh medium with the same composition. Among all combination of growth hormones 1.0 mg/l BAP and 0.1 mg/l NAA gave highest number of shoot proliferation than other hormone concentration (Table 1). At higher concentration of BAP few number of shoot were proliferated.

The acclimatization and field establishment work is in research. It was observed that *In vitro* propagate *Gmelina arborea* plants from explants showed that multiplication rate are still low and leaves were defoliated therefore research to improve the protocol efficiency is to be continued.

This table show that explants responded to all media. Media with BAP 1.0 mg/l and NAA 0.1mg/l showed best results for shoot initiation from nodal explant. Each nodal explant with single node gives rise 2-4 shoot after 4 week. When subculture in high concentration of BAP 3.0 mg/l showed explant swelling only but no any shoot proliferation was obtained. The best concentration for shoot multiplication and condition of plant was found in hormone concentration 1.0 mg/l BAP and 0.1 mg/l NAA. In this composition, micro shoots were very healthy and strong.
Discussion

There are only few research work carried out for micropropagation of *Gmelina arborea*. Nakamura (2006) used shoots apices as an explant and culture on ½ Gamberg media solidify with 3.2 gm/l of gelrite and he used growth hormone 0.0 and 0.002 mg/l of IBA and sucrose. Induced multiple shoots were subculture into same media for micropropagation of *Gmelina arborea*. Lisette Valverde-Cerdas, Laura Alvarado, Ana Hine (2004) inoculated nodal segment of germinated seedling in different concentration of Benzylamino purine. Isidro E. Suarez, Claudia C, Acosta, Kellen C, Gatti (2013) found that control of BAP showed increase in shoot proliferation, shoot length and leaf number while IBA induce higher root number, root length and rooting percentage. Higher survival rate were observed when plants were transfered to peat.Gamboa & Abdelnour (1999) reported a new shoot formation from explants with pre-existing meristems, isolated from *in vitro* germinated plants, cultured in 2.46ìM BAP supplied MS semisolid medium. Kannan V.R. & Jasrai Y.T. (1996), obtained multiple shoots from single node explants of mature *Gmelina arborea* Roxb. on MS medium supplemented with 6-benzyladenine (BA). Seven to nine shoots were formed when in vitro derived single node explants were subcultured on MS medium supplemented with 1.1 micromolar BA. Most of the researcher used nodal cutting as an explant for micropropagation of *Gmelina arborea*. They used growth hormone Benzyl amino purine and Naphthalene acetic acid for microshoot regeneration. In our research work we used nodal explant as an initial plant material for micropropagation of *Gmelina arborea*. Incase of growing media we used MS media with growth hormone Benzyle amino purine and Naphthalene acetic acid. We found low concentration of auxin was suitable and condition of plants was also found to be healthy.

Conclusion

*Gmelina arborea* Roxb. is multipurpose tree which is cultivated in many country including Nepal. Tissue culture is only the best tool for true to type plant production. The objective of this present research work was to maximize the number of microshoots per culture bottle manipulating the appropriate concentration of growth hormones. We found Benzyl amino purine (BAP) 1.0 mg/l and Naphthalene acetic acid (NAA) 0.1 mg/l promoted higher frequency of shoot proliferation.

Acknowledgement

It is our pride privilege to express our sincere thanks and deep sense of gratitude to Director General Mr. Rajdev Prasad Yadav, Deputy Director General Ms. Susma Upadhya and Mr. Sanjeev Kumar Rai for facilitating lab making it suitable to conduct this research successfully.

References


http:www.Forestrynepal.org resources/trees/ Gmelina arborea

Nakamura, K. 2006. Micropropagation of *Shorea roxberghii* and *Gmelina arborea* by shoot apex culture. Plantation technology in tropical forest science, 137-150

Clonal propagation of *Gmelina arborea* Roxb. by Nodal culture

**Figure 1:** Nodal explant culture

**Figure 2:** Shoot regeneration after 4 weeks

**Figure 3:** 6 weeks old micro plants
Endemic Flowering Plants of Nepal: An update

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Abstract

The existing list of the endemic flowering plants of Nepal is revised, the present list of the flowering plants of Nepal is updated on the basis of published literature and the status of these endemic plants is presented. The result shows that there are 324 species of Nepalese endemic flowering plants distributed under 132 genera and 45 families.

Key words: Endemic plants, flowering plants, flora of Nepal, endemism, Himalaya.

Introduction

Endemic flowering plants of Nepal are those plants whose distributions are confined to Nepal. If the plant is found outside Nepal then it is not endemic to Nepal. In a wider sense, the distribution of any endemic taxonomic unit is confined to a particular country or geographical region (Good, 1974; Rajbhandari, 2013). The high mountains and islands have shown greater endemism in general. The high number of endemic species in such area is attributed to the isolation that allows the persistence and divergence of the population (MacArthur & Wilson, 1967; Bruchmann & Hobohm, 2014). Importance of the endemic plants lies on the fact that if it is lost from a particular country or region it will be lost from the world as well. Therefore, utmost importance has to be given for the conservation of these plants.

The first information on the endemic flowering plants of Nepal was provided by Hara & Williams (1979) and Hara et al. (1978, 1982) in their books ‘An enumeration of the flowering plants of Nepal volumes 1-3’ which later became a source for preparing the list of the endemic flowering plants of Nepal. Joshi and Joshi (1991) provided a list of 283 endemic flowering plants of Nepal giving emphasis on their conservation. Shrestha & Joshi (1996) reported 246 species of endemic flowering plants of Nepal of which 77 species have now been reported from outside Nepal and 155 species have been added to the list as endemic to Nepal (see Appendix 1 to 4). On the basis of the list of Shrestha & Joshi, Chaudhary (1998) has also given a list of endemic flowering plants of Nepal. Ghimire (2005) reported endemic flowering plants of Dolpo (northwest Nepal) area. Bhuju et al. (2007) in ‘Nepal biodiversity resource book: Protected areas, Ramsar sites and world heritage sites’ have included as an Annex 1.5 a list of the endemic flowering plants and have recorded 316 species out of total 399 taxa (species, subspecies, variety) including some common species distributed in India, Nepal, Bhutan and China, e.g., Habenaria edgeworthii Hook. f. ex Collett, Oberonia pratitiana King & Pantl., Peristylus densus (Lindl.) Satapau & Kalipada, and Poa rajbhandarii Noltie. Out of 316 species listed 100 species have been recorded from outside Nepal and are henceforth no more endemic to Nepal. The Department of Plant Resources published endemic flowering plants of Nepal in three parts which included 282 species (Rajbhandari & Adhikari, 2009; Rajbhandari & Dhungana, 2010, 2011a). Rajbhandari & Dhungana (2011b) and Rajbhandari (2013) analysed the diversity of Nepalese endemic flowering plants and noted that the families having large number of endemic species were Asteraceae (22 species), Saxifragaceae (21 species), Papaveraceae (20 species) and Ranunculaceae (20 species) and the genera having large number of endemic plants were Saxifraga (21 species), Pedicularis (12 species), Meconopsis (11 species) and Impatiens (10 species).
species). Rajbhandari et al. (2015) reported that in the National Herbarium of Nepal (KATH) herbarium specimens of 128 species of endemic flowering plants of Nepal are preserved. Miehe et al. (2015) have noted that 399 endemic flowering plants of Nepal listed by Bhuju et al. (2007) are unevenly distributed. The number of endemic flowering plants of Nepal is not yet clearly known. An update is now necessary to make clear the status of the endemic flowering plants of Nepal. The objective of this paper is to present the status of the endemic flowering plants of Nepal by revising their existing list and updating them on the basis of published literature.

Results

Among 282 species of endemic flowering plants of Nepal reported by the Department of Plant Resources in the books ‘Endemic flowering plants of Nepal part 1 to 3’ 18 species have been reported from outside Nepal and 60 species have been added as endemic to Nepal (see Appendix 2, 3). In total, occurrence of 132 species (2.4%) of the previously recorded Nepalese endemic flowering plants has been reported from outside Nepal (Appendix 1).

The present list includes 324 species of Nepalese endemic flowering plants distributed under 132 genera and 45 families (Table 1, Appendix 4). This is about 6% of the 5500 species of flowering plants recorded from Nepal (Rajbhandari, 2016). Twelve flowering plant families in Nepal have 10 or more than 10 endemic species (Table 2). Among them the families having large number of endemic species are Apiaceae (28 species), Asteraceae (22 species), Fabaceae (21 species), Saxifragaceae (21 species), Orchidaceae (20 species), Papaveraceae (20 species) and Ranunculaceae (20 species). Sixteen genera have 5 or more than 5 species of endemic flowering plants in Nepal (Table 3). The genera having large number of endemic plants are Saxifraga (21 species), Pedicularis (13 species), Meconopsis (11 species), Aconitum (10 species) and Impatiens (10 species). There is no endemic flowering plant family in Nepal. However, there is one endemic flowering plant genus (Discretithecaceae) in Nepal. Discretithecaceae belonging to Lamiaceae family was first described by P. D. Cantino in an article written by Cantino et al. in 1999. This is a monotypic genus which contains only one known species Discretithecaceae nepalensis (Moldenke) P. D. Cantino. Nine tree species are found among the endemic plants of Nepal. These species are Mallotus bicarpellatus T. Kuros., Litsea doshia (D. Don) Kosterfer., Machilus pubescens Blume, Salix nepalensis Yonek., Prunus himalaica Kitam., Prunus jajarkotensis H. Hara, Prunus taplejungnica H. Ohba & S. Akiyama, Prunus taplejungnica H. Ohba & S. Akiyama and Sorbus sharmae M. F. Watson, V. Manandhar & Rushforth.

Table 1. List of endemic flowering plants of Nepal with families and genera.

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<thead>
<tr>
<th>Family</th>
<th>Genera</th>
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</thead>
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<td>Lalldhwojia – 2 species</td>
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<tr>
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<td>Oreocome – 2 species</td>
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<td>Pimpinella – 3 species</td>
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<tr>
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<td>Rohmooa – 1 species</td>
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<tr>
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<td>Sinocarum – 2 species</td>
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<td>Synclinostyles – 2 species</td>
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<td>Vicatia – 1 species</td>
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<tr>
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<td>Conioselinum – 1 species</td>
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<tr>
<td></td>
<td>Cortia – 1 species</td>
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<tr>
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<td>Cortiella – 1 species</td>
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<td></td>
<td>Dolpojestella – 1 species</td>
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<tr>
<td></td>
<td>Heracleum – 1 species</td>
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<tr>
<td></td>
<td>Justicia – 1 species</td>
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<td>Strobianthes – 3 species</td>
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<td>Thunberga – 1 species</td>
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<td>Allium – 1 species</td>
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</tr>
</tbody>
</table>
Asteraceae
Artemisia – 2 species
Cicerbita – 1 species
Cirsium – 2 species
Crepis – 1 species
Leontopodium – 2 species
Saussurea – 7 species
Senecio – 2 species
Synotis - 2 species
Taraxacum – 3 species

Balsaminaceae
Impatiens – 10 species

Begoniaceae
Begonia – 7 species

Berberidaceae
Berberis – 2 species

Boraginaceae
Arnebia – 1 species
Microula – 1 species
Onosma – 3 species

Brassicaceae
Aphragmus – 2 species
Draba – 3 species
Lepidostemon – 1 species
Noccaea – 1 species
Solms-laubachia – 2 species

Campanulaceae
Codonopsis – 3 species
Cyananthus – 2 species

Caryophyllaceae
Arenaria – 2 species
Silene – 7 species

Crassulaceae
Rhodiola – 1 species
Rosularia – 1 species
Sedum – 1 species

Cucurbitaceae
Gomphogyne – 1 species

Cyperaceae
Carex – 7 species

Elaeagnaceae
Elaeagnus – 1 species

Ericaceae
Rhododendron – 2 species

Eriocaulaceae
Eriocaulon – 4 species

Euphorbiaceae
Croton – 1 species
Leptopus – 1 species
Mallotus – 1 species

Fabaceae
Astragalus – 9 species
Colutea – 1 species
Crotalaria – 1 species
Hedysarum – 1 species
Millettia – 1 species
Oxytropis – 7 species
Rhynchosia – 1 species

Gentianaceae
Gentiana - 5 species
Gentianella – 2 species
Swertia - 3 species

Hypericaceae
Hypericum – 1 species

Iridaceae
Iris – 1 species

Juncaceae
Juncus – 2 species

Lamiaceae
Clinopodium – 1 species
Discretitheca – 1 species
Eriophytan – 2 species
Isodon – 3 species
Microtoena – 1 species
Nepeta – 1 species
Salvia – 1 species

Lauraceae
Litsea – 1 species
Machilus – 1 species
Lythraceae
*Rotala* – 1 species

Oleaceae
*Jasminum* – 1 species

Onagraceae
*Epilobium* – 2 species

Orchidaceae
*Bhutanthera* – 1 species
*Bulbophyllum* – 1 species
*Eria* – 4 species
*Gastrochilus* – 1 species
*Herminium* – 1 species
*Liparis* – 2 species
*Malaxis* – 2 species
*Neottia* – 2 species
*Oberonia* – 1 species
*Odontochilus* – 1 species
*Oreorchis* – 1 species
*Panisea* – 1 species
*Pleione* – 1 species
*Sunipia* – 1 species

Orobanchaceae
*Euphrasia* – 1 species
*Pedicularis* – 13 species

Papaveraceae
*Corydalis* – 9 species
*Meconopsis* – 11 species

Plantaginaceae
*Lagotis* – 1 species
*Veronica* – 1 species

Poaceae
*Borinda* – 1 species
*Elymus* – 1 species
*Eulaliopsis* – 1 species
*Festuca* – 3 species
*Himalayacalamus* – 5 species
*Poa* – 3 species

Saccharum – 1 species
*Sinarundinaria* – 1 species
*Stipa* – 1 species
*Thamnocalamus* – 1 species

Polygonaceae
*Bistorta* – 3 species
*Fagopyrum* – 1 species
*Fallopia* – 1 species

Primulaceae
*Primula* – 5 species

Ranunculaceae
*Aconitum* – 10 species
*Anemone* – 1 species
*Clematis* – 3 species
*Delphinium* – 3 species
*Oxygraphis* – 1 species
*Ranunculus* – 2 species

Rosaceae
*Potentilla* – 2 species
*Prunus* – 4 species
*Sibbaldia* – 1 species
*Sorbus* – 1 species

Rubiaceae
*Galium* – 2 species
*Ophiiorrhiza* – 1 species

Salicaceae
*Salix* – 3 species

Saxifragaceae
*Saxifraga* – 21 species

Scrophulariaceae
*Scrophularia* – 2 species

Urticaceae
*Pilea* – 1 species

Zingiberaceae
*Roscoea* – 3 species
Table 2. List of families having ten or more than ten species of endemic flowering plants of Nepal

<table>
<thead>
<tr>
<th>Family</th>
<th>Species</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apiaceae</td>
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<td>Balsaminaceae</td>
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<td>Lamiaceae</td>
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Table 3. List of genera having five or more than five species of endemic flowering plants of Nepal

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
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</thead>
<tbody>
<tr>
<td>Saxifraga</td>
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<tr>
<td>Pedicularis</td>
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<tr>
<td>Meconopsis</td>
<td>11</td>
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<tr>
<td>Aconitum</td>
<td>10</td>
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<tr>
<td>Impatiens</td>
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<tr>
<td>Astragalus</td>
<td>9</td>
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<tr>
<td>Corydalis</td>
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<tr>
<td>Acronema</td>
<td>8</td>
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<tr>
<td>Begonia</td>
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<td>Carex</td>
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<td>Oxytropis</td>
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<td>Saussurea</td>
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<td>Silene</td>
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<td>Gentiana</td>
<td>5</td>
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<tr>
<td>Himalayacalamus</td>
<td>5</td>
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<tr>
<td>Primula</td>
<td>5</td>
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</table>

Conclusion

For the proper conservation measures to be undertaken for the endemic flowering plants of Nepal a systematic investigation of these plants should be carried out in order to understand their natural habitat, which will furnish the ecological requirements of different species. Rajbhandari et al. (2015) reported that there are 128 species of herbarium specimens of endemic flowering plants of Nepal preserved in the National Herbarium of Nepal (KATH). For the identification as well as to know the locality and habitat of these endemic plants it is essential that the specimens of all the Nepalese endemic species should be represented in the Herbarium of Nepal.

Acknowledgements

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References


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Appendix 1: List of the flowering plants of Nepal reported as endemic to Nepal by different authors (Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Bhuju et al., 2007; Rajbhandari & Adhikari, 2009; Rajbhandari & Dhungana, 2010, 2011a; Rajbhandari et al., 2015), but now also reported from outside Nepal. (Distr. = Distribution. W. = West Nepal (from western border to 83° E longitude. C. = Central Nepal (from 83° E longitude to 86°30’ E longitude). E. = East Nepal (from 86°30’ E longitude to eastern border).

**ACANTHACEAE**

**Strobilanthes angustifrons** C. B. Clarke in Hooker, Fl. Brit. Ind. 4: 466 (1884).


Distr.: India (West Himalaya), W. Nepal.


Distr.: C. Nepal, China (Tibet).


*Ruellia rotundifolia* D. Don, Prodr. Fl. Nepal. 120 (1825).

Distr.: India, C. Nepal, Bhutan, China.

**Strobilanthes tamburensis** C. B. Clarke in Hooker, Fl. Brit. Ind. 4: 454 (1884).

Distr.: E. Nepal, Bhutan.


*Aechmanthera tomentosa* Nees in Wallich, Pl. Asiat. Rar. 3: 87 (1832).


Distr.: Pakistan, India, Nepal, Bhutan, Bangladesh, China, Myanmar, Laos.

**APIACEAE**


**Cortiella hookeri** (C. B. Clarke) C. Norman, J. Bot. 75: 94 (1937).

**Cortia depressa** (D. Don) C. Norman, J. Bot. 75: 96 (1937).


**Cortia nepalensis** C. Norman, J. Bot. 67: 245 (1929).

**Cortia lindleyi** DC., Prodr. 4: 187 (1830).

**Cortia oreomyrrhiformis** Farille & Malla, Candollea 40: 545 (1985).

Distr.: Nepal, India (Darjeeling, Sikkim), Bhutan, China (Tibet).


Distr.: C. Nepal, Bhutan.

Distr.: C. Nepal, Bhutan.

*Peucedanum glaucum* DC., Prodr. 4: 179 (1830).
Distr.: India, C. Nepal.

Distr.: C. Nepal, China (Tibet).

Distr.: C. Nepal, India (Darjeeling).

**Pleurospermum rotundatum** (DC.) C. B. Clarke in Hooker, Fl. Brit. Ind. 2: 703 (1879).
*Hymenolaena rotundata* DC., Prodr. 4: 245 (1830).
Distr.: C. Nepal, China (Tibet).

**APOCYNACEAE**

Distr.: India, W. Nepal.

**ARACEAE**

**Arisaema costatum** (Wall.) Mart. ex Schott, Melet. Bot.: 17 (1832).
Distr.: C. Nepal, China.

Distr.: E. Nepal, China.

Distr.: E. Nepal, China (Tibet).

**ASTERACEAE**

Distr.: C. Nepal, Bhutan.

Distr.: Nepal, India (Darjeeling).

Distr.: C. Nepal and China (Tibet).

Distr.: Nepal and China (Tibet).

Distr.: E. Nepal, India (Sikkim).

Distr.: C. Nepal, India (Sikkim), Bhutan.

**Saussurea graminifolia** Wall. ex DC., Prodr. 6: 536 (1838).
Distr.: India, Nepal, Bhutan, China.

Distr.: E. Nepal, China.

Distr.: W. Nepal, India (E. Himalaya).

**BALSAMINACEAE**

**Impatiens insignis** DC., Prodr. 1: 688 (1824).
Distr.: C. Nepal, India (E. Himalaya).

**Impatiens leptoceras** DC., Prodr. 1: 688 (1824).
Distr.: C. Nepal, India (E. Himalaya), China.

Distr.: E. Nepal, Bhutan.
BERBERIDACEAE

Berberis hamiltoniana Ahrendt, Gard. Ill. 64: 426 (1944).
Distr.: India, Nepal, China (Tibet).

Distr.: India, W. Nepal.

Berberis wallichiana DC., Prodr. 1: 107 (1824).
Distr.: C. Nepal, India (E. Himalaya), Bhutan.

BRASSICACEAE

Distr.: C. Nepal, India (Sikkim).

Diplotaxis harra (Forssk.) Boiss., Fl. Orient. 1: 388 (1867).
Distr.: SW Europe, N Africa, SW Asia, Afghanistan, Pakistan, India, W. Nepal.

Distr.: India, C. Nepal, Bhutan, China, Myanmar.

Distr.: India, Nepal, China, Myanmar, Cambodia, Vietnam.

Distr.: C. Nepal, China.

CELASTRACEAE

Distr.: India, Nepal, China, Myanmar, Cambodia, Vietnam.

CYPERACEAE

Distr.: C. Nepal, India (Sikkim).


Scirpus juncoides Roxb., Fl. Ind. 1: 218 (1820).

ERIOCAULACEAE


EUPHORBIACEAE


FABACEAE


GENTIANACEAE

Gentiana decemfida Buch.-Ham. ex D. Don, Prodr. Fl. Nepal.: 127 (1825). Distr.: India (Himalaya), Nepal.


LAMIACEAE


Distr.: W. Nepal, China (Tibet).

**LENTIBULARIACEAE**

Distr.: E. Nepal, India (Sikkim).

**ORCHIDACEAE**


Distr.: Pakistan, India, Nepal, Bhutan.

Distr.: India, Nepal.


Distr.: Nepal, India (Sikkim).

**OROBANCHACEAE**


Distr.: India, W. Nepal, Bhutan, China.

Distr.: C. Nepal, China (Tibet).

*Pedicularis siphonantha* D. Don, Prodr. Fl. Nep.: 95 (1825).

Distr.: India, W. Nepal, Bhutan, China.


Distr.: C. Nepal, China (Tibet).

**PAPAVERACEAE**


Distr.: India (Uttar Pradesh, Sikkim), W. Nepal, Bhutan.

*Corydalis latiflora* Hook. f. & Thomson, Fl. Ind.: 270 (1855).

Distr.: W. Nepal, India (Sikkim), Bhutan, China (Tibet).


Distr.: W. and C. Nepal, India (Sikkim), Bhutan.

Distr.: C. & E. Nepal, China (Tibet).

**POACEAE**

Distr.: C. Nepal, China (Tibet).


Distr.: C. Nepal, China (Sichuan, Yunnan).


Distr.: C. Nepal, Bhutan, China.


*Elymus microlepis* Melderis in Bor, Grass. Ind.: 692 (1960).


**POLYGONACEAE**


**PRIMULACEAE**

**Primula aureata** Fletcher, Gard. Ill. 63: 283 (1941). Distr.: C. and E. Nepal, India (Sikkim).

**RANUNCULACEAE**


**Delphinium altissimum** Wall., Pl. Asiat. Rar. 2 (6): 25 (1831). Distr.: C. Nepal, India (NE India), Bhutan, China (Tibet).


**Thalictrum dalzellii** Hook., Icon. Pl. 9: t. 868 (1852). Distr.: India, Nepal.

**Thalictrum rotundifolium** DC., Syst. Nat. 1: 185 (1817). Distr.: C. Nepal, China (Tibet).


**Sibbaldia adpressa** Bunge in Ledebour, Fl. Altaic. 1: 428 (1829).

Distr.: Russia, C. Nepal, China, Mongolia.

**RUBIACEAE**

**Rubia alata** Wall. in Roxburgh, Fl. Ind. 1: 384 (1820).
Distr.: C. Nepal, China.

**Wendlandia puberula** DC., Prodr. 4: 412 (1830).
Distr.: India, C. Nepal, Myanmar.

**SALICACEAE**


**Blackwellia napaulensis** DC., Prodr. 2: 54 (1825).
Distr.: India, Nepal, China.

Distr.: E. Nepal, China.

Distr.: C. Nepal, India (Sikkim), China.

**SCROPHULARIACEAE**

Distr.: C. Nepal, India (Sikkim).

**ZINGIBERACEAE**

**Roscoea capitata** J. E. Sm., Trans Linn. Soc. 13: 461 (1822).
Distr.: C. Nepal, China (Tibet).
Appendix 2. The following 19 species which were recorded as endemic to Nepal by Rajbhandari & Adhikari (2009), Rajbhandari & Dhungana (2010, 2011a) and Rajbhandari et al. (2015) are now non-endemic to Nepal due to their reports of occurrence outside Nepal.

**ACANTHACEAE**


*Aechmanthera tomentosa* Nees in Wallich, Pl. Asiat. Rar. 3: 87 (1832).


Distr.: Pakistan, India, Nepal, Bhutan, Bangladesh, China, Myanmar, Laos.
Ref.: Hara et al., 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.

**APIACEAE**


*Peucedanum glaucum* DC., Prodr. 4: 179 (1830).

Distr.: India, C. Nepal.
Ref.: Hara et al., 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.

**APOCYNACEAE**


Distr.: India, W. Nepal.
Ref.: Hara et al., 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.

**ASTERACEAE**

*Saussurea graminifolia* Wall. ex DC., Prodr. 6: 536 (1838).


Distr.: India, Nepal, Bhutan, China (Tibet).


**ERIOCAULACEAE**


Distr.: E. Nepal, India (Himalaya).

**EUPHORBIACEAE**


Euphorbia schillingii Radcl.-Sm. in Kew Mag. 4 (3): 112 (1987).

Distr.: Nepal, India (Sikkim), Bhutan, China.

Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**LAMIACEAE**


Distr.: E. Nepal, Bhutan.


Distr.: W. Nepal, China (Tibet).


**PAPAVERACEAE**


Distr.: C. & E. Nepal, China (Tibet).

Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2010.

**POACEAE**


Distr.: C. Nepal, China (Tibet).


Distr.: India (Garhwal), C. Nepal.

Ref.: Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2010.

**RUBIACEAE**

Rubia alata Wall. in Roxburgh, Fl. Ind. 1: 384 (1820).

Distr.: C. Nepal, China.

Ref.: Hara et al., 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.

Wendlandia puberula DC., Prodr. 4: 412 (1830).


Distr.: India, C. Nepal, Myanmar.

Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.
Appendix 3. The following 60 endemic species of Nepal are not recorded in the *Endemic Flowering Plants of Nepal part 1, 2, 3* and *Catalogue of Nepalese flowering plants Supplement 1* (Rajbhandari & Adhikari, 2009; Rajbhandari & Dhungana, 2010, 2011a; Rajbhandari et al., 2015).

**ACANTHACEAE**

*Strobilanthes nutans* ((Nees) T. Anders.  
*Goldfussia nutans* Nees  
*Thunbergia nepalensis* Bh. Adhikari & J. R. I. Wood

**APIACEAE**

*Acronema bryophilum* Farille & Lachard  
*Acronema cryptosciadeum* Farille & Lachard  
*Acronema mukherjeeanum* Farille & Lachard  
*Acronema phaeosciadeum* Farille & Lachard  
*Acronema pneumatophobium* Farille & Lachard  
*Acronema refugicolum* Farille & Lachard  
*Conioselinum nepalense* Pimenov & Kljuykov  
*Lalldhwojia pastinacifolia* Pimenov & Kljuykov  
*Oreocome depauperata* Pimenov & Kljuykov  
*Oreocome involucellata* Pimenov & Kljuykov  
*Pimpinella acronemastrum* Farille & Lachard  
*Pimpinella kawalekhensis* Farille & Lachard  
*Rohmooa kirmzii* Farille & Lachard  
*Sinocarum staintonianum* P. K. Mukherjee ex Farille & Lachard  
*Synclinostyles denisjordanii* Farille & Lachard  
*Synclinostyles exadversum* Farille & Lachard

**ASTERACEAE**

*Synotis managensis* S. Joshi, K. Shrestha & D. Bajracharya

**BALSAMINACEAE**

*Impatiens recticalcarata* S. Akiyama

**BEGONIACEAE**

*Begonia nuwakotensis* S. Rajbhandary  
*Begonia panththarensis* S. Rajbhandary  
*Begonia taligera* S. Rajbhandary

**BERBERIDACEAE**

*Berberis pendryi* Bh. Adhikari

**BRASSICACEAE**

*Lepidostemon williamsii* (H. Hara) Al-Shehbaz  
*Draba williamsii* H. Hara

**CUCURBITACEAE**

*Gomphogyne nepalensis* W. J. de Wilde & Duyfjes

**CYPERACEAE**

*Carex staintonii* X. F. Jin, H. Ikeda & O. Yano

**EUPHORBIACEAE**

*Leptopus nepalensis* B. Adhikari, R. P. Chaudhary & S. K. Ghimire

**FABACEAE**

*Astragalus jumlaensis* Podlech  
*Astragalus lobbichleri* Podlech  
*Astragalus nepalensis* Podlech  
*Astragalus notabilis* Podlech  
*Astragalus poluninii* Podlech  
*Astragalus pseudorigidulus* Podlech

**ORCHIDACEAE**

*Bhutanthera fimbriata* B. B. Raskoti  
*Bulbophyllum nepalense* Raskoti & Ale  
*Gastrochilus nepalensis* B. B. Raskoti  
*Herminium hongdeyuanii* B. B. Raskoti  
*Liparis langtangensis* B. B. Raskoti & Ale  
*Neottia chandrae* B. B. Raskoti, J. J. Wood & R. Ale  
*Odontochilus nandae* Raskoti & H. Kurzweil  
*Panisea panchaseensis* Subedi  
*Sunipia nepalensis* B. B. Raskoti & R. Ale

**OROBANCHACEAE**

*Pedicularis yamazakiana* R. R. Mill

**PAPAVERACEAE**

*Corydalis simplex* Liden  
*Meconopsis autumnalis* P. A. Egan  
*Meconopsis lamjungensis* T. Yoshida, H. Sun & Grey-Wilson  
*Meconopsis manasluensis* P. A. Egan  
*Meconopsis simikotensis* Grey-Wilson

**POACEAE**

*Himalayacalamus planatus* Stapleton  
*Poa hideaki-ohbae* Rajbh.  
*Sinarundinaria langtangensis* R. Manandhar &
Bajracharya

RANUNCULACEAE
Clematis staintonii W. T. Wang
Delphinium unifolium Tamura

ROSACEAE
Prunus taplejunica H. Ohba & S. Akiyama,
Prunus topkegolensis H. Ohba & S. Akiyama

RUBIACEAE
Galium nepalense Schoenb.-Tem. & Ehrendorfer
Galium saipalense Schoenb.-Tem. & Ehrendorfer
Ophiorrhiza nepalensis Deb & Mondal

SALICACEAE
Salix staintoniana Skvortsov

**ACANTHACEAE**

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: W. Nepal.

**AMARYLLIDACEAE**

Distr.: C. Nepal.

**APIACEAE**

Distr.: C. Nepal.

Distr.: E. Nepal.

Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: E. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: E. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.


Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.

Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: W. Nepal.

Distr.: W. Nepal.

Distr.: C. Nepal.


Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: W Nepal

Distr.: C. Nepal. 


Distr.: C. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: W. Nepal.
Ref.: Press et al., 2000; Rajbhandari & Dhungana, 2011.

APOCYNACEAE


Distr.: W. Nepal.
Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. Nepal.

ASPARAGACEAE

Distr.: W. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

ASTERACEAE

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: E. Nepal.

Distr.: W. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: W. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. Nepal.
Balsaminaceae

Distr.: E. Nepal.

Distr.: W. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: C. Nepal.

Distr.: E. Nepal.

Distr.: C. Nepal.

Distr.: W. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. & C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**BEGONIACEAE**

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: E. Nepal.
Ref.: Hara & Williams, 1979; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: E. Nepal.

Distr.: C. Nepal.

Distr.: E. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**BERBERIDACEAE**

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

**BORAGINACEAE**


**Macrotomia nepalensis** Kitam. in Kihara, Peoples Nep. Him. 423 (1957).
Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**Microula mustangensis** Yonek. in Ohba et al., Fl. Mustang, Nepal: 244 (2008).
Distr.: C. Nepal.

Distr.: W. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.


Distr.: E. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.


**Maharanga wallichiana** A. DC., Prodr. 10: 71 (1846).
Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**BRASSICACEAE**


Distr.: E. Nepal.


Distr.: W. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: W. Nepal.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.


Distr.: W. & C. Nepal.

Distr.: W. & C. Nepal.

**CAMPANULACEAE**

Distr.: C. Nepal.


Distr.: C. Nepal.

*Cyananthus hayanus* Marq., New Fl. & Silv. 8: 207 (1936).
Distr.: C. Nepal.
Ref.: Hara et al., 1982; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.
Distr.: C. Nepal.

**CARYOPHYLLACEAE**

*Stellaria mukerjeeana* Majumdar, Blumea 16: 267 (1968).
Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: W. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

**Silene helleboriflora** Exell & Bocquet, Candollea 17: 37 (1959).
Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.


Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**Silene vautierae** Bocquet, Candollea 22: 17 (1967).
Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**CRASSULACEAE**

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

**CUCURBITACEAE**

Distr.: C. Nepal.
CYPERACEAE

Distr.: E. Nepal.

Distr.: C. Nepal.

Distr.: E. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

Distr.: C. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.

ELAEAGNACEAE

Distr.: C. Nepal.

ERIIOCAULACEAE

Distr.: E. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: C. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: E. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

Distr.: E. Nepal.
Ref.: Hara et al., 1978; Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Adhikari, 2009.

EUPHORBIACEAE

Distr.: C. Nepal.
Distr.: C. Nepal.

Distr.: C. Nepal.

**FABACEAE**

Distr.: C. Nepal.

Distr.: W. Nepal.

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**Astragalus lobbichleri** Podlech, Feddes Repert. 120(1&2): 50 (2009).
Distr.: C. Nepal.

Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rahbhandari & Dhungana, 2010.

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Distr.: C. Nepal.
Ref.: Hara & Williams, 1979; Shrestha & Joshi, 1996; Press et al., 2000; Rahbhandari & Dhungana, 2010.

**Millettia nepalensis** R. N. Parker, Kew Bull. 1931: 42 (1931).
Distr.: W. Nepal.
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**GENTIANACEAE**

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Distr.: C. Nepal.
Ref.: Hara et al., 1982. Press et al., 2000;
Rajbhandari & Dhungana, 2010.

Distr.: E. Nepal.

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**Gentianella glanduligera** Airy Shaw in Hooker, Ic. Pl. 35: t. 3431 (1943).
Distr.: C. Nepal.
Ref.: Hara et al., 1982. Press et al., 2000;
Rajbhandari & Dhungana, 2010.

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Ref.: Hara et al., 1982. Press et al., 2000;
Rajbhandari & Dhungana, 2010.

Distr.: E. Nepal.

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**HYPERICACEAE**

**Hypericum cordifolium** Choisy in DC., Prodr. 1:
IRIDACEAE

Distr.: C. Nepal.

JUNCACEAE

Distr.: C. Nepal.

Distr.: C. Nepal.

LAMIACEAE


Distr.: C. Nepal.


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Ref.: Shrestha & Joshi, 1996; Rajbhandari & Dhungana, 2010.


Distr.: C. Nepal.


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Distr.: E. Nepal.
Ref.: Shrestha & Joshi, 1996; Rajbhandari & Dhungana, 2010.


Distr.: W. and C. Nepal.


_Rabdosia phulchokienensis_ Murata in Fl. E. Him. 3: 96 (1975).
Distr.: C. Nepal.

Distr.: E. Nepal.

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LAURACEAE

Distr.: C. and E. Nepal.

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LYTHRACEAE

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ONAGRACEAE

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**Pedicularis odontoloma** Yamazaki, J. Jap. Bot. 53:


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Distr.: C. Nepal.


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Ref.: Hara & Williams, 1979; Press et al., 2000; Rajbhandari & Dhungana, 2010.

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Meconopsis napaulensis DC., Prodr. 1: 121 (1824).

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Agropyron nepalense Melderis in Bor, Grass. Ind.: 692 (1960).
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**Saccharum williamssii** (Bor) Bor, Grasses Burma, Ceylon, India & Pakistan: 214 (1960).

**Erianthus williamssii** Bor, Kew Bull. 1957: 413 (1957).
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**Stipa staintonii** Bor, Bull. Bot. Surv. Ind. 7: 133 (1965).


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Distr.: C. Nepal.

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Ref.: Shrestha & Joshi, 1996; Press et al., 2000; Rajbhandari & Dhungana, 2011a.

**ROSACEAE**

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Distr.: E. Nepal.

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Ref.: Hara & Williams, 1979; Shrestha & Joshi,


RUBIACEAE


SALICACEAE


SAXIFRAGACEAE


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URTICACEAE

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Department of Plant Resources. 2007. Medicinal plants of Nepal (Revised), *Bull. Department of Plant Resources No. 28*, Thapathali Kathmandu, Nepal.


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