



# Smithsonian Environmental Research Center

June 1, 2021

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Dear Mgr. Havrdová

As requested by email on May 17, 2021, I am providing the following evaluation of the chapters of a Ph.D. thesis submitted by Binu Timsina. I have also mailed a copy of the evaluation.

## **Overall Evaluation**

Binu Timsina has presented a thesis composed of nine chapters based on research in Nepal. Six of the chapters have been published in peer-reviewed journals or conference proceedings. The remaining three chapters have been submitted for publication. In addition to individual chapters, Timsina provided a summary of her thesis that included background information on the overall goals of her research, results obtained from each component of the research and recommendations for future efforts based on her research. Timsina also provided a description of her involvement with the topic of each chapter, demonstrating that she was the primary motivator of each project and was responsible for data collection and analysis and manuscript preparation.

When working in a country that is still, in part, in the alpha-stage of knowing what species are present, the thesis demonstrates a broad approach that includes efforts to quantify orchid species richness in the country, to an assessment of distribution factors and the likely impact of climate change on a one orchid species. Given the important of ethnobotanical use of native orchids, Timsina also engaged in efforts to better understand the uses of medicinal plants, including how they are used and their chemical composition. Finally, Timsina conducted a 7-year demographic study of an important orchid species.

The last three chapters are not focused on orchids but link the orchid-based research to the broader issue of ethnobotanical uses of plants in Nepal. As described below, the final three chapters are also valuable contributions.

The thesis demonstrates a high degree of motivation and an ability to develop, conduct and complete a thesis in a part of the world that is often resource limited. The compilation of products is clearly worthy of a Ph.D and the results of Timsina's efforts represent a valuable contribution to orchid conservation globally as well as within Nepal.

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It is also noteworthy that Binu Timsina is Nepalese and her first four degrees were taken in Nepal. She began her Ph.D. research in the Czech Republic five years after completing her M.Sc. in Nepal. Her CV demonstrates that she remained active between the time the M.Sc. was completed and the onset of the Ph.D. program. Since matriculating in the Ph.D., she has been very active, giving presentations and publishing manuscripts that are not part of the thesis.

### **Comments on specific chapters.**

**Chapter 1.** An annotated checklist of the orchids of Nepal. Published in the *Nordic Journal of Botany*

The goal of this effort was to solve nomenclatural problems and ambiguity existing for various species, and update the list of orchids published till present in Nepal. Timsina was the 3<sup>rd</sup> author of this contribution. Results of the effort are presented in a checklist format resulting in documentation for 458 taxa. The Method section demonstrates that the approach to compiling information on orchids in Nepal was rigorous; including a literature survey, field collections and information gathered from herbaria. The publication is an important contribution to the literature and will provide a basis for future research on orchids of Nepal.

**Chapter II:** Orchids of Nepal: phytogeography and economic importance. Publishes as a chapter in *Global Change: A Complex Challenge* published by the Global Change Research Centre

Timsina is the first author of this multi-authored contribution. This short (4 page) contribution is a logical extension of the information compiled in Chapter 1. The authors assign the known orchid taxa to four physiographic areas and quantified the similarity of the orchid flora between the areas.

The contribution could have been more robust if the authors had expanded the Discussion and analysis to focus on specific issues such as the endemics and why there were large differences between some of the regions. With more effort, this could have been a contribution in a peer-reviewed journal.

**Chapter III.** Diversity, distribution and host-species associations of epiphytic orchids in Nepal. Published in *Biodiversity Conservation*.

Timsina is the lead author of this multi-authored contribution. Epiphytes were the focus of the field-based sampling effort in five locations. The authors had four goals: "(1) does the diversity of epiphytic orchids in different localities of Nepal differ? (2) What is the relationship between the abundance of host species and the number of orchid species living on these hosts? (3) What are the factors determining presence, diversity and composition of orchids on particular host plants? (4) How do these associations vary in different localities?"

The study was conducted over several years and almost 23,000 trees were sampled for epiphytes. Characteristics of the trees (e.g., bark texture, evergreen or not) were obtained and used in the analyses.

They found a rich diversity of orchid epiphytes and the pattern of distribution and abundance were found to be related primarily to the abundance of host trees, the characteristics of the host and, to a degree, the plant families of the trees. They also found differences between localities.

This will be a valuable contribution to individuals and organizations that focus on the distribution of epiphytic orchids and the factors that relate to the abundance and where they occur. To date, this is one of the most rigorous studies of this sort. I found the relationship between epiphyte diversity and the abundance of host trees the most interesting result.

**Chapter IV.** Associations between epiphytic orchids and their host plants and their future in Nepal in the context of global warming. Submitted by no journal designation indicated.

This contribution is a follow-up to Chapter II. Data compiled for Chapter III were further analyzed to identify specific associations between orchids and host trees and then use climatic data to project what might happen in response to global warming of 5 C.

Three questions were addressed: 1. Which host species are more likely to host multiple epiphytic orchid species on them?, 2. What is the pattern of distribution of all host and epiphytic orchids along altitudinal gradients?, 3. How many species of host and epiphytic orchids are potentially threatened by global warming, based on the projected increase in temperature?

The Phi coefficient of association was used to determine the relationship between host trees and epiphytic orchids. Other analytical efforts were similar to those described in Chapter III.

It would have been useful to have a more thorough description of the procedures that were used to link tree species distribution to temperature and how that was linked to projected temperature changes. Specifically, I did not see a link between the description of how data were processed that resulted in Figure 7.

The Discussion is well written, but several issues are not considered that might be important. The authors indicated that animals may disperse faster than plants. What is the basis for this, especially for orchids that have wind dispersed seeds and may be poised to disperse those seeds to a wide range of potential host trees? Which comes first, the tree or the orchid? Changes in orchid abundance might be independent, to some degrees, from their tree hosts as they also have independent life history characteristics that need to be considered. How will the orchids respond to climate change? The focus of the Discussion is the tree host.

**Chapter V.** Epiphytic orchid diversity along an altitudinal gradient in central Nepal. Accepted for publication in *Plants*.

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Timsina is the lead author of this multi-authored peer reviewed manuscript.

This contribution is an attempt to provide more insight into the distribution of orchids on host trees. As in the other contributions in the thesis, altitude is an important factor but, in this study, consideration was also given to other potentially important factors such as soil pH, precipitation and the distance of the host tree from the forest edge.

Precipitation data were downloaded from a web site. How was it calibrated to the scale of each tree (or altitudinal zone) sampled?

How was the soil sampled to get a pH measurement? Was it a composited from several samples of soil collected around the tree? What depth of soil was represented in the sample that was analyzed?

The ordination (Figure 3) shows pattern but I wonder how much confidence you can have in discussing them because they explain so little of the variance?

An interesting outcome of study was the notation that finer-scale sampling resulted in finding more orchids, some not previously known from the local flora.

While altitude was the most important factor, the author recognized and described limitation of the study and the need for other studies to identify the proximate factors responsible for the presence or absence of orchids on tree hosts.

**Chapter VI.** Six-year demographic study of the terrestrial orchid, *Crepidium acuminatum*: implications for conservation. Submitted for publication.

This chapter represents a six year demographic study of a terrestrial orchid. Matrix modelling was used to compare population growth with different regimes of herbivory and harvesting. The value of the research is that it provides scenarios for harvesting the species that can result in sustainable populations.

The authors were able to adequately assess the dynamics of small plants that likely emerged from seeds. It was not possible to determine from the methods section of the new plants were from seeds or from asexual propagules.

**Chapter VII.** Xanthenes content in *Swertia multicaulis* D. Don from Nepal. Published in *Molecules*.

This chapter, for which Timsina is the lead author, is about a non-orchid species that is in a Genus that is known to have medicinal uses. The phytochemistry of species that was examined had not been previously evaluated. The species is harvested for medicinal uses in Nepal.

The research was not based on testing any specific hypothesis, but the information gained is useful because it identifies the levels of Xanthonenes in a species that had not been previously assayed.

The chapter did not contain a Discussion of the meaning of the results, other than they concluded that it was very rich in xanthone content.

**VIII. *Neopicrorhiza scrophulariiflora* (Pennell) Hong:** A comprehensive review of its traditional uses, phytochemistry, pharmacology and safety. Published in *Journal of Ethnopharmacology*.

Timisina is the 4<sup>th</sup> or a four-authored publication that is a compilation of information on the uses, phytochemistry, and pharmacology of a species in the Plantaginaceae. Based on the characterization of the species, including the uncertain taxonomic status and wide uses, a review of the species is a valuable contribution.

The contribution is based completely on review and evaluation of available data for the species.

I don't have the knowledge to evaluate the results of the review but based on the extent and thorough evaluation of existing data, I believe that this is an important contribution. In addition, the authors provide a list of topics that need further evaluation – thus presenting a framework for additional research.

**Chapter IX. Traditional uses of medicinal plants in gastrointestinal disorders in Nepal.** Published in *Journal of Ethnopharmacology*.

Timisina is the 4<sup>th</sup> author in this 9-authored publication. The focus is a review of medicinal plants in Nepal that are used for gastrointestinal disorders.

The study was a compilation of existing information followed by a meta-analysis of the data. They evaluated 947 species in 158 plant families and 586 genera, including orchids.

Similar to Chapter VII, I can't evaluate the pharmacological aspects of the study but the science is nicely presented and evaluated. The Discussion is thorough, and the authors provide a framework for additional research that links the ethnobotanical uses of the species to conservation issues.

Sincerely



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