The Rufford Foundation
Final Report

Congratulations on the completion of your project that was supported by The Rufford Foundation.

We ask all grant recipients to complete a Final Report Form that helps us to gauge the success of our grant giving. The Final Report must be sent in **word format** and not PDF format or any other format. We understand that projects often do not follow the predicted course but knowledge of your experiences is valuable to us and others who may be undertaking similar work. Please be as honest as you can in answering the questions – remember that negative experiences are just as valuable as positive ones if they help others to learn from them.

Please complete the form in English and be as clear and concise as you can. Please note that the information may be edited for clarity. We will ask for further information if required. If you have any other materials produced by the project, particularly a few relevant photographs, please send these to us separately.

Please submit your final report to jane@rufford.org.

Thank you for your help.

Josh Cole, Grants Director

Grant Recipient Details	
Your name	Tashi Yangchen
Project title	Conifers Conservation in Himalayan Country: Diversity and Infection Assessment of Plant Parasitic- Mistletoes in Conifer Forests in Thimphu District of Bhutan
RSG reference	20248-1
Reporting period	Sept 2016 to Oct 2017
Amount of grant	£4996
Your email address	tyang2121993@gmail.com
Date of this report	14/10/2017

1. Please indicate the level of achievement of the project's original objectives and include any relevant comments on factors affecting this.

Objective	Not achieved	Partially achieved	Fully achieved	Comments
1. **Assessment of the mistletoe infection at on the conifer forest species.**				**Study area** Thimphu is divided into four administrative ranges, Gidakom, Thimphu, Khasadrupchu and Chamgang Ranges, but the study was focused in Gidakom, Thimphu and Khasadrupchu Ranges. **Forest Types Identification** Topographic maps of study area (scale 1:50,000) were scanned and transferred to ERDAS IMAGINE 8.7 for geo-referencing. Satellite image of study area was extracted from IRS-1D-LISS-III from USGS site or Bhuvan sites and was rectified with topographic map of study area in ERDAS IMAGINE. After rectification subset of AOI (area of interest) was classified into different land uses (agriculture, forest, barren land, and water) and different forest class (fir forest, conifer forest, mixed forest, broadleaved forest and scrub forest) using the non-supervise classification in ERDAS IMAGINE to be used in cluster random sampling. **Cluster (Area) Sampling** Thimphu district was divided into three ranges (Thimphu Range, Kasadrupchu Range and Gidakom Range) and were considered as separate area. In each range with use of remote sensing and GIS forest was classified into different forest class which was called as clusters (smaller area). The ground truthing was performed with the help of Forest Division officials of Thimphu. A total of 35 sampling units (40 x 40 m) were drawn randomly and framed datasheets were used to study diseases in clusters having tree

				species of interest. Different conifers forest class (only conifers forest class) according to size of it, numbers of sampling unit (40 x 40 m) was drawn randomly and framed datasheets was used to study diseases (mistletoe infection) in sampling units. Cluster sampling was opted because study area was large and needed to be subdivided into smaller areas for the convenience and random sampling for overcoming non-probability sampling. Cluster random sampling, gives equal probability of selecting all conifer trees within its population in Thimphu district. It reduces bias in site selection to be included in the sample. As a result, the cluster random sample provides with a sample that is highly representative of the population being studied. Following formula will be used to calculate with number of sampling units required. N1=C1/TA Where, N1=Maximum number of sampling units needed in one forest class C1= Size of conifer forest class (m) TA= Size of sampling plot (m) Or, N=C/TA N= Maximum number of sampling units needed in project area C= Total area of conifer forest in project area (m) TA= Size of sampling plot (m) **Calculation** According to Norbu *et al.* (2010), the blue pine coverage in Thimphu is 187 km^2, broadleaf is 122 km^2, broadleaf with conifer is 5 km^2, Chir pine is 28 km^2, fir is 118 km^2 and mixed conifer is 388 km^2 in the Thimphu district. Thimphu has approximately 726 km^2 conifer forests coverage. According

				to the literature review, mistletoes are commonly found on the blue pine. Hence only blue pine forest was considered. Total conifers forest of Thimphu districts= 726 km^2 Average area of study area= 726~700 km^2 However, only blue pine forest coverage of 187 km^2 was considered. Size of sampling plot is 40 x 40 m or (1600 m^2) Therefore, number of sampling unit required =187,000m^2/1600m^2=116 sampling units. By taking 30% of sampling intensity=30/100*116=35 sampling units **Sampling** Sampling plot area has major influence on the sampling intensity, time and resources spent in the field measurements. Therefore, sampling plots size of 1600 m^2 (40 x 40 m) was used and 35 sample plots were laid by using cluster random sampling method. **Data collection methods** **Mistletoe Infection Assessment** 3.3.4.1.1 Survey team members The team members comprised of atleast four persons; two persons for measuring the DBH (diameter at breast height) and height of tree, one person who was good at plant identification, and one person to measure elevation, slope and aspect. **Field equipment** Parameters such as, GPS coordinates, slope gradient, elevation, aspects, girth and height of the trees were measured. GPS coordinates and elevation of the sample plots were measured by GPS device. Slope gradient and aspects of the sample plots were measured by clinometer and compass respectively. The girth and height of the individual trees

				were measured using DBH tape meter and digital hypsometer. **Mistletoe infection rating (DMR method)** DMR system was used to assess the severity of mistletoe infection in the conifers forests. **1. Site of infestation** Site of infestation refers to the occurrence of the mistletoe on the tree. According to the site of infestation occurring on the tree, it was categorized as stem, branches, and stem and branches. **2 Part of infestation** Part of infestation refers to occurrence of the mistletoe on the part of the tree. The tree was visually divided into upper, middle and lower part. The severity of infection was recorded based on its infection on the part of the tree. **3 Severity of mistletoe infection** Infection severity refers to the intensity of infection caused by mistletoe on the host plant. In most conifer species, the severity of mistletoe was quantified using the Six-Class Draft DMR System. This system was based on the proportion of mistletoe-infected branches in each infected tree. This method provides a quantitative reference scale for determining the relative population status of a mistletoe infestation within a stand, its potential for spread and intensification and helps to calculate the disease severity index (DSI). This DSI incorporates all the DMR classes and provides a simple, synthetic summary in a single index. It is standard for the quantifying the mistletoe infection rate. This helps in predicting and forecasting the mistletoe infection in conifer forests and take necessary preventative measures. The 6-Class DMR system method

<table>
<tr><td></td><td></td><td></td><td></td><td>(Hawksworth, 1977) was used for rating the mistletoe infection in conifer trees. Infected trees were divided visually into three parts and each part were be rated “0”for no mistletoes infection, “1” for light mistletoe infection (Less than half of branch infected) and “2” for heavy infection (More than half of branch infected). Rating form each part was added to obtain a total rating for the tree. [(Ratings class 0 (uninfected tree), 1-2 (light), 3- 4 (moderate), and 5- 6 (severe)].
Rating Class
Hawsworth’s “dwarf mistletoe rating system (DMR)”, where ratings range from 0 to 6 developed by Hawsworth was used for the assessment of the mistletoe infection (Hawksworth and Wiens, 1996). After the addition of the infection severity of the respective part of the tree, rating class were developed in which ratings class 0 for uninfected tree, 1-2 for light mistletoe infection , 3- 4 for moderate mistletoe infection and 5- 6 for severe infection.
Confirmation of mistletoe species
Mistletoe infection on trees was based on observed symptoms on affected trees (branches, twigs and stems). The samples were collected and photographed from the field. The herbarium were prepared for the scientific identification of mistletoe from National Biodiversity Centre, Bhutan and authentication from Herbarium of Systematics Branch of Botany and Forest Pathology Division of Forest Research Institute, Dehradun, India with the expertise of the supervisor and taxonomists.</td></tr>
<tr><td>2. To assess mistletoe infection at different site condition such as aspects, elevation and slope gradient.</td><td></td><td></td><td></td><td>Following are the site condition which were taken into account:
<table><tr><td>Infection assessment variables</td><td>Sampling methods</td></tr></table></td></tr>
</table>

				Tree species composition	Enumerate all plant species of >1.37 m height as trees within the sampling unit
				DBH of tree	A plant of height >1.3 m was measured by diameter tape.
				Tree height	Height measured by Digital hyposometer
				Elevation	Elevation was recorded from the center of sample plot by altimeter
				Aspect	Determined aspect from the center of sample plot by compass as N, NE, E, SE , S, SW, W, NW
				Slope	Measured slope in degrees from the center of the sample plot
3. Determine the diversity of mistletoe in conifer forest species in Thimphu district.				**Mistletoe severity index and incidence** According to method of Mathiasen *et al.* (2008), disease severity index (DSI) in each conifer species was calculated from the sum of total number of trees of each disease severity rating (DSR) in all the plots multiplied separately by the disease class (1 - 6) and dividing it by the total number of trees assessed (N) as given in the following formula. **3.3.4.6.2 Infection/Disease severity index** Disease severity is the measure of degree of damage done by mistletoe on the host plants. DSI = (nC1 x 1 + nC2 x 2 + ….. C6 x6)/ (N *Maximum disease class (6)) *100 Where: nCl, nC2…nC6 =Total number of trees in each infection rating class 1,2,3…..6 is the severity rating class of	

				mistletoe infection. N = Total number of trees assessed in all the observation plots. **Mistletoe incidence** Mistletoe incidence is the number of host plants infested by mistletoe within a pollution. The per cent incidence of mistletoe was calculated from the total number of plants affected (nd) and total number of plants observed in all the plots (N): Mistletoe incidence = nd/N*100 **Biodiversity index of mistletoes** Biological diversity can be quantified in many different ways. The two main factors taken into account when measuring diversity are richness and evenness. Richness is a measure of the number of different kinds of organisms present in a particular area. Evenness is a measure of the relative abundance of the different species making up the richness of an area. Diversity has remained a central theme in ecology and is seen as an indicator of the wellbeing of ecological system. The diversity takes into account the number of species and how the species are equally abundant. Mistletoes diversity in each host was tested using the Shannon-Wiener index (1949) and Simpson's index. These indexes incorporate both components of biodiversity (evenness and richness) and it provides a simple, synthetic summary in a single index $\textbf{Shannon index } (\mathbf{H}') = -\sum pi \ln pi$ Where, H'=the Shannon diversity index *pi*= S/N, fraction of the entire population made up of species *i* *S* = numbers of species encountered N= Total number of all individuals in the sample *lnpi* = natural log of *pi* Simpson's Diversity Index is a measure of diversity. In ecology, it is often used

				to quantify the biodiversity of a habitat. It takes into account the number of species present, as well as the abundance of each species. **Simpson's diversity index D**=1-$\sum p^2 i$ S=Number of individual of one species N=Total number of all individuals in the sample
Estimate the effect of mistletoes on economic value of conifer tree species.				**Economic value** Information on number of species affected (measure DBH and height of each infected trees) was used to estimate mistletoe impact on economic value. Using the local volume prepared by the Ministry of Agriculture and Forest of Bhutan, DBH of infected and un-infected were compared with the standard volume of the individual tree. As per Hawksworth and Geils (1996), a light infection does not have a measurable impact on tree or stand growth, but a heavy infection can reduce potential volume to as little as one-seventh, or 14 %, that of a healthy stand. Hence it is obvious that it affects the productivity. According to severity intensity rating class, loss of volume is calculated by 14% loss of its production. Then the loss is multiplied with the standard cost of timber given by Natural Resource Pricing Committee (NRPC) of Bhutan. **Calculation** Actual volume of each tree= DBH of individual tree* Standard volume of the DBH range from volume table Total volume production: Sum of actual volume of each tree Loss of volume production: 14% of total volume production (Hawksworth and Geils, 1996). Actual loss expressed in money: Loss of volume production *standard cost of the timber

2. Please explain any unforeseen difficulties that arose during the project and how these were tackled (if relevant).

It was very difficult to identify and locate the presence of *Arceuthobium minutissimum* with the naked eyes from far as it being very minute. It shows multiple primary branching and emerges from basal cups, which remain embedded in the bark, when shoots are shed. Himalayan dwarf mistletoe is an extremely reduced plant and lacks secondary branching. And it was very difficult to locate its presence. However, Taxillus kaempferi was easily identified. Binoculars were used so that minute *Arceuthobium minutissimum* is identified easily.

Dwarf mistletoe rating system (DMR)", where ratings range from 0 to 6 developed by Hawsworth in the early 1950's was used to quantify the infection intensity of mistletoe on conifer forest species. In the 6-class system, the live crown is visually divided into three equal parts, and each third is rated as 0 for no mistletoe visible, 1 for light mistletoe infection (less than half of the branches infected), or 2 for heavy mistletoe infection (more than half of the branches infected). Ratings for each third are then added to obtain a total for the tree. As a result there were chances of bias in the assessment. However same person took the responsibility of quantifying so that chances of bias are reduced. An assessment of DMR was done using binoculars and it was based on the presence of symptoms and signs of the disease, especially brooms and aerial shoots of dwarf mistletoes.

The harsh climatic condition was another challenge. The heavy monsoon rain in summer and varied topographical gradient (rugged and steep slopes) with strong wind and cold weather was one of the challenges. Some of the study sites were very far from the road and travelling was also one of the difficulties.

3. Briefly describe the three most important outcomes of your project.

Knowledge base
This project aimed to study mistletoe diversity and their threat assessments on conifers will be valuable reference for a scientific protection and management of conifers forest. It will serve as the base line information and assist the conservation planners to consider the role of mistletoe play in the conservation approach. It helped students, foresters, locals and policy makers aware that not only wildfire and anthropological destruction but also pathogens are threats to the forest conservation through presentation and publication of project findings. It helps to make people clear on the concepts of healthy forest for the vibrant ecosystems and habitats for rich biodiversity. Hence, this study helped to give the scope of forest pathogens in detrimental effect of conifer forests to conservationist for scientific management and planning.

Diversification of conservation measures
This project is justified to detect parasitic mistletoe in which the early detection will allow timely eradication or control action, minimize the losses of conifer value and limit the cost of control. Hence this will facilitate the diversification of the conifers protection and conservation approach to forest pathology apart from traditional

social system, wild fire and anthropological activities. This project also aimed to impart conifers conservation approach by integrating traditional and scientific knowledge with changing time.

Ecology and economic
Mistletoe causes loss in productivity of conifer forest species. Shanon-Weiner diversity and Disease Severity Index which provide a simple and synthetic summary will provide the valuable information for analyzing the economic loss, ecological stability, planning, control and scientific management of mistletoe in conifers forests.

4. Briefly describe the involvement of local communities and how they have benefitted from the project (if relevant).

Large group of people were involved during the entire project period as one of the project objectives is participatory based. My team consists of following people who helped me for the successful completion of this project:

1. One forestry student (B.Sc.) - as Field Assistant in different sites for collecting data and monitoring of the project.
2. One contractual labourers - for performing various tasks like collection of pathogens (mistletoe), infected samples from field, etc.
3. Foresters (two) - since the sites falls under Thimphu district field surveillance was done by foresters as regular assigned duties and field guide in the study area.
4. For the identification and assessment, Ngawang Dema (M.Sc. Forestry), Choney Yangzom (M.Sc. Forestry, Phurpa M.Sc. Forestry) of Forest Research Institute , Dr. Karma Wangchuck (Taxonomist of Sherubtes college), Ugyen Tshering (M.Sc. Forestry, India Forest Service), Dr. N.S.K. Harsh (Forest Pathology Division, FRI) and the Systematic Branch of Forest Research Institute, India and National Biodiversity Centre, Bhutan. Infection rate and other assessments was helped by the Forest Pathology Division, of FRI, India.

Major Involvement took place while conducting conservation awareness campaign and stakeholders workshop including involving Department of Forest and Part Services, National Biodiversity Centre, Royal Society of Protection of Nature (RSPN), Ugyen Wangchuck Institute for Conservation and Environment (UWICE), students of Royal Thimphu college and forestry trainees of College of Natural Resources (CNR) from Bhutan and MSc Scholars of Forest Research Institute from India, have actively participated and provided opportunity to discuss and share their opinions on impact of mistletoe on conifer forest species. Creative and programmatic suggestions were suggested to improve on the methodology and additional areas to be covered were discussed. Refreshments were also served to the participants.

Involvement of local community in awareness campaign took place with the help of local leaders who have huge influence in the community. People were local people had achieved basic information about the kind of species found in their locality and mistletoe impact on the forest. Both local leader and local people who involved in

the project were financially benefited as they were provided daily payment as per local rate as wages.

5. Are there any plans to continue this work?

Conifer tree species are one of the most economically important tree species where Bhutanese depends for various raw materials such as timber, firewood, etc. But mistletoe cause economic loss of productivity of the conifer forest species. It is seen important to have a long term monitoring in their habitats. Hence, Department of Forest supports the project and detection parasitic mistletoe in which the early detection will allow timely eradication or control action, minimize the losses of conifer value and limit the cost of control.

6. How do you plan to share the results of your work with others?

To outreach the sustainability in conservation field, the good knowledge and experiences gained from current project including the main project findings are to be shared to research enthusiast, conservationist, focal research institutions and organizations. So far, results of the project were disseminated as mentioned below:

1. Results and information were presented to final year students of BSc Sustainable Development at College of Natural Resources and also to the students of Royal Thimphu College.

2. Research finding was presented to conservation NGOs at World Wildlife Fund (WWF Bhutan program) conference hall where seven research focal persons from RSPN and WWF Bhutan were gathered. Results of the project were further shared by circulating the same presentation slides which I have submitted to RSGF.

3. Findings and results were presented as my MSc Thesis to the MSc Forestry students and PhD scholars of Forest Research institute (Deemed) University, Dehradun, India.

4. Further, the results and information will be submitted to Ugyen Wangchuck Institute for Conservation and Environment (UWICE), National Biodiversity Centre (NBC) and Department of Forest and Park Services (DoFPS).

7. Timescale: Over what period was The Rufford Foundation grant used? How does this compare to the anticipated or actual length of the project?

The grant was fully used for a period of 12 months as project commenced from August 2016 to July 2017 as same timescale proposed in the project. However, I had to make little adjustment with proposed time scheduled depending on weather condition, capture success and convenience.

Following are activities in which grant were used:

1. Stakeholders workshop involving Department of Forest and Part Services, National Biodiversity, Royal Society of Protection of Nature (RSPN), Ugyen Wangchuck Institute for Conservation and Environment (UWICE) from Bhutan and Forest Research Institute from India, would be organized before the implementation of the project in October, 2016.
2. One-day field training (theory and practical) for project team and one forest personal on sampling (site selection), method of sample collection, equipment handling, safety measure and distribution of safety wears before onset of field work
3. In November and December 2016 first round of sample collection in Thimphu Forest Range of Thimphu district.
4. Results and information were presented to final year students of BSc Sustainable Development at College of Natural Resources and also to the students of Royal Thimphu College.
5. Research finding was presented to conservation NGOs at World Wildlife Fund (WWF Bhutan program) conference hall where seven research focal persons from RSPN and WWF Bhutan were gathered. And fund was used while creating awareness to the local community.
6. Since it was my MSc I had to finish the data collection before the completion of my course. Therefore the second field data was collected in April and May, 2017.
7. Findings and results were presented as my MSc Thesis to the MSc Forestry students and PhD scholars of Forest Research institute (Deemed) University, Dehradun, India in end of July, 2017.

8. Budget: Please provide a breakdown of budgeted versus actual expenditure and the reasons for any differences. All figures should be in £ sterling, indicating the local exchange rate used.

Item	Budgeted Amount	Actual Amount	Difference	Comments
Field equipment's - Measuring tape, vernier calliper, GPS, altimeter, binocular, digital camera, compass, pocket calculator, stopwatch tents	905	875	+30	All the material were purchased from the online shopping of India: www.flipcart.com and www.amazon.com
Travel cost	450	455	-5	Vehicle hiring charge was higher in remote villages due to high maintenance costs.
Standard field kits/stationaries (Printer paper, field notebook,	50	55	-5	Two field guide book for identifying tree species were purchased.

field guide book, lead pencils, plastic bag, ruler)				
Communication	310	200	+110	Mobile phones and email/internet.
Education and Awareness	1020	1100	-80	The refreshment was provided during the entire awareness program and workshop with the forest official, conservationist during the discussion of finding and further improvement or protection program to be doing in the area.
Budgets for printing Poster, Broachers and Pictorial	200	205	-5	Posters were used during local visit in the community and during presentation aside slide presentation.
Budget for salaries/wages	2062	1989	+73	The wages to the team member were calculated based on the as monthly salaries.
Total	4997	4879	118	

9. Looking ahead, what do you feel are the important next steps?

1. Studying the distribution and diversity of different types of mistletoe in western, Bhutan.
2. Identifying the factors which support the growth of mistletoes plants on the host.
3. Studying the mode of mistletoe's dispersal in conifer forests.
4. Community based conservation with involvement of local people, students and related stake holders.

10. Did you use The Rufford Foundation logo in any materials produced in relation to this project? Did The Rufford Foundation receive any publicity during the course of your work?

With an aim to publicise RF to conservationists, students, local community, focal research institution and organizations, I have used the Rufford Foundation logo while making presentations to:

1. The final year BSc Sustainable Development students of College of Natural Resources, Lobesa.
2. The District Forest Officer, Extension Forest Officers and local people of Thimphu District (study area). By this time, majority of the people in study area can recognize the RSGF logo.
3. The Rufford Foundation logo was used while conducting training on survey techniques and skills and presentations to the students of Royal Thimphu College.

4. The logo was also used during field visit while creating awareness to the people.
5. RF logo will be used in upcoming presentation scheduled on 14th June, 2017 at Forest Research Institute (Deemed) University, Dehradun, India. Further, RF logo will be used in all the materials produced from this project and most importantly, RF will be highly acknowledged in relevant publications of this study in future.

11. Please provide a full list of all the members of your team and briefly what was their role in the project.

The team consists of following:

1. One forestry student (B. Sc.) - as Field Assistant in different sites for collecting data and monitoring of the project.

2. One contractual labourer - for performing various tasks like collection of pathogens (mistletoe), infected samples from field, etc.

3. Foresters (2) - since the study falls under Thimphu district field surveillance is done by foresters as regular assigned duties and will act as field guide in the study area.

4. Dr. N.S.K. Harsh, Scientist -G (Retired), Forest Pathology Division, Forest Research Institute, Dehradun, India, Loday Phuntsho, Deputy Chief Research Officer, Renewable Natural Resources Research and Development Centre, Wengkhar Ministry of Agriculture and Forests, Mongar, Bhutan and Mr. Tshering Phuntsho, Researcher and Coordinator of Conservation & Sustainable Livelihoods Program, Royal Society for Protection of Nature (RSPN), Kawajangsa, Thimphu, Bhutan were my referees and they guided me till the end .

5. In field identification and assessment, Ngawang Dema (M.Sc. Forestry), Choney Yangzom (M.Sc. Forestry, Phurpa M.Sc. Forestry) of Forest Research Institute were key resource personnel. Karma Wangchuck (Taxonomist of Sherubtes College), Ugyen Tshering (M.Sc. Forestry, India Forest Service) and the Systematic Branch of Forest Research Institute, India and National Biodiversity Centre, Bhutan, helped me with the identification of herbarium. Infection rate and other assessments was helped by the Forest Pathology Division, of FRI, India as I was attached to this Division. Kinley Dorji (M.Sc. Environment Management), Sangay Tshering (M.Sc. Environment Management), and Karma Sherub (M.Sc. Forestry) of Forest Research Institute, (Deemed) University, Dehradun, India, also helped me with the research.

12. Any other comments?

With a heartfelt appreciation, I would like to thank your esteemed RF for providing kind financial grant, without which I could not have captured a single bat.

To keep the good knowledge and experiences gained from this project alive, and to progress the bat conservation work in near future, I look forward to a similar financial support from your esteemed foundation. So with deep reverence, I would like to request your esteemed foundation to look into the consideration to reopen and continue granting support to Bhutan with some formalities. The continuation grant from your esteemed foundation would make vast contribution in nature conservation projects.