Conservation collections at the National Botanic Garden of Nepal – the Franklinia Taxus Project

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Abstract

Three species of *Taxus* occur in Nepal: *T. contorta*, *T. mairei* and *T. wallichiana*. All are under pressure from collection for medicinal use, from habitat destruction and from changing land use. In 2017 and 2018 fieldwork, funded by Fondation Franklinia, was carried out to collect seed, herbarium specimens, cuttings and information to improve protection of these species. Collections were made for propagation, herbarium and molecular studies. Population-level information was also gathered to make national conservation assessments for these species. This article traces the status of *Taxus* spp. in the living collections in the National Botanic Garden of Nepal (NBG) and the conservation assessments made by the team comprising staff of the Department of Plant Resources (DPR) and the Nepal Academy of Science and Technology (NAST), as well as a freelance botanist. Reference is made to work done prior to 2017 on the taxonomy and distribution of the species and which enabled efficient fieldwork thereafter. The establishment of a conservation collection at NBG provides a basis for research into yew species in Nepal and highlights the benefits of internationally collaborative projects to maintain conservation collections of threatened species in their country of origin.

Introduction

The conservation of species is dependent upon understanding the status of, and threats to, species in the wild. Living collections of threatened species in the protected environment of a botanic garden can substantially support conservation efforts. Living material in such conservation collections can be used for research, education and commercial farming, and to augment wild populations when conditions are right.

A two-year collaborative project that ran from 2017 to 2019 entitled 'Conservation of *Taxus contorta* and building capacity for IUCN Red Listing in Nepal', funded by Fondation Franklinia through Botanic Gardens Conservation International (BGCI), was undertaken by the Royal Botanic Garden Edinburgh (RBGE) and the Department of Plant Resources (DPR), Ministry of Forests and Environment of the Government of Nepal. The focus species was *T. contorta* Griff. (West Himalayan yew), and a number

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of activities brought in the two other threatened yews that occur in Nepal, *T. wallichiana* Zucc. (East Himalayan yew) and *T. mairei* (Lemée & Lév.) S.Y.Hu ex T.S.Liu (Maire's yew). The aims of the project were to complete a survey of the populations of all three in West and Central Nepal, update the knowledge base of *Taxus* in Nepal, build capacity for *ex situ* conservation and strengthen *in situ* conservation. An additional aim was to deliver training in IUCN conservation assessment and to produce IUCN assessments for these three species (Watson, 2017).

Fondation Franklinia is a private foundation established in 2005 under Swiss law. Its objective is to preserve threatened tree species throughout the world and improve their conservation status, and it provides grants to support nature conservation projects (Fondation Franklinia, 2000–2022). To ensure that all tree species are assigned a threat category, the foundation also supports the Global Tree Assessment, a monumental initiative led by BGCI and the Global Tree Specialist Group of the IUCN Species Survival Commission to assess all 60,000+ tree species by 2023 (BGCI, 2021).

Taxus is one of the most threatened genera in the Hindu Kush Himalaya region of which Nepal is part (Poudel *et al*., 2012). Throughout the region, harvesting foliage for anti-cancer compounds and local use for medicine, fuel, fodder and construction have negatively impacted up to 90 per cent of wild populations (Poudel *et al*., 2012).

Poudel *et al*. (2012) established that three clearly delineated species of yew occur in Nepal: *Taxus contorta*, *T. mairei* and *T. wallichiana*. In 2013 they highlighted the ethnographic relevance of yews in the context of traditional uses as practised by major ethnic groups inhabiting Nepal from west to east (Poudel *et al*., 2013). All three species are used by local people. Bhatt *et al.* (2017) conducted extensive work on the distribution of the three species. Some trees form part of community-managed woodlands which are regularly cut for commercial collection of leaves to produce the cancer drug Paclitaxel. Many trees in the national forests are cut illegally for timber and firewood and, as in West Nepal, debarked to make local herbal tea (Fig. 1). The health of the populations of all three species is impacted.

Habitat reduction and disturbance due to grazing and development for agriculture and for road building have an impact on the biodiversity of any area, including on that of yews. In addition, when larger tree species such as *Tsuga dumosa* (Himalayan hemlock) are cut for construction and fuel wood the primary canopy under which yew species are adapted to grow disappears and exposes the shade-adapted secondary canopy to full sun. Higher exposure to sun causes scorching on the leaves and dries the soil out, reducing vigour and preventing healthy shoot growth (Fig. 2). *Taxus mairei* is particularly affected because it occurs at lower elevations where pressure on land is greater and human population numbers are higher (Bhatt *et al*., 2017).

The Franklinia Taxus Project set out to explore further the health and status of some of the main populations in Nepal, collect material with which to establish an *ex situ* living collection at the National Botanic Garden of Nepal (NBG) in Godavari, Lalitpur, and complete Red List assessments for all three species. The project's final activities comprised two training visits to the UK in October 2019 carried out by Nepali researchers.

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Fig. 1 *Taxus contorta* debarked for use in tea. Photo: Kate Hughes.



Fig. 2 An ageing specimen of *Taxus contorta* exposed to high light levels and low soil moisture following removal of the primary canopy. Photo: Kate Hughes.

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Methods

Fieldwork and collections

Collections were made on two field trips, one in October–November 2017 and the other in December 2018. Both visits were timed to coincide with expected seeding time of *Taxus* species. Living and herbarium specimens and information were collected of every population accessed. A distance of 1 km or more between trees observed was considered to merit collection. At each population the geographic extent was estimated and the health of the population documented. This included recording the forest type, associated tree and shrub species, signs of regeneration, size and number of mature yew trees, ratio of male to female trees, impact of grazing and other human-related activities such as felling of trees, cutting of branches, evidence of fire and other disturbances. Whenever possible, local people, including Community Forest User Groups, were asked for their perceptions of changes in the condition of the forests and the known use of the forests (Fig. 3).

Herbarium, leaf and cuttings collections were made from one tree in each population only (Fig. 4). Details were taken on the dimensions, health, sex and location, including coordinates, of the tree sampled. Associated species, the aspect (north or south) and factors affecting the growing environment and tree health, such as the presence of grazing animals or landslides, were noted. Typical foliage and fruiting stems of male and female representatives were selected for herbarium specimens, and leaf material was collected in tea bags and placed in a box with colour-changing silica gel.

Each collection was given a number preceded by the collection code FTP (for Franklinia Taxus Project). Strong, clean and healthy epicormic growth was prioritised for cuttings, but if this was not available material was taken from healthy branches.



Fig. 3 Consultation with Community Forest User Groups at Chulipran Community Forest. Photo: Kate Hughes.

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Fig. 4 Collection of material from *Taxus contorta* at Bulbule, Jumla, western Nepal. Photo: Mark Watson.

Epicormic growth is known to result in plants with greater apical dominance and which therefore form trees with a better shape than those taken from lateral branches (M. Gardner, pers. comm.). Cuttings material was taken from both male and female trees within a population. A long-handled pole pruner was used to reach high branches. Material for herbarium specimens was labelled and dried in a standard herbarium press. Leaf material collected in paper bags for DNA extraction was transferred to an airtight box containing fresh silica gel to dry it out efficiently.

Cuttings material was labelled, formed into a compact bundle and wrapped in damp cotton cloth, then placed in food preparation bags formulated to keep food fresh for longer than conventional polythene bags (Fig. 5). The bags are trademarked Stayfresh™ and are available from Lakeland.[5] They were stored in conditions that were as cool as possible, out of strong sunlight, and checked regularly (Fig. 6).

Seed was put into labelled cloth drawstring bags and kept cool and dry. Ripe, or nearly ripe, seeds were collected where available. Where there was a paucity of ripe seed, nearly-red fruit was collected and left to ripen in labelled and sealed plastic bags. The ideal seed for sowing is black with a fleshy red aril around it. On immature fruits both seed and aril are green and the colours change as the seed ripens over several weeks (Fig. 7).

Herbarium voucher specimens with associated collection data and digital images were collected for all samples taken, supplemented with silica-dried leaf material for DNA analysis (Fig. 8). Specimens and dried leaf material were deposited in the national herbarium of Nepal (KATH). Cuttings and seed were deposited with NBG. Both institutions

[5] www.lakeland.co.uk

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Fig. 5 Bags of cuttings material and seed processed for storage before their arrival at NBG. Photo: Kate Hughes.



Fig. 6 Cuttings material was checked regularly and kept as cool as possible. Photo: Kate Hughes.

are governed by the DPR, and both are located at Godavari, 16 km south-east of the capital city of Kathmandu.

Cultivation of collections

Living collections were established at NBG following each field trip using the cuttings material and seed collected. On both occasions training was given to NBG horticultural staff in the management of protected environments for rooting cuttings of woody species, preparing seed for sowing and maintaining plant records for conservation collections.

Vegetative propagation

Cuttings were taken to NBG and immediately processed. The material was prepared and divided into labelled bundles, then soaked in a solution of indolebutryic acid

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Fig. 7 Fruiting shoot and seeds of *Taxus contorta* collected from Depalgaun, Jumla, western Nepal. The shoot shows fruits at different stage of ripening. Photo: Kate Hughes.



Fig. 8 Herbarium specimens were collected from each tree sampled. Photo: Kate Hughes.

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(IBA). Once soaked for the recommended time, the cuttings were inserted into open ground and protected with cloches. IBA and recommended treatments were sourced from the Rhizopon® website.[6]

This was the first time that this method of taking cuttings had been used at NBG. Seven employees, including horticultural managers and nursery staff, were given training in its use, including processing records and taking semi-ripe stem cuttings (Figs 9 & 10). Some of the equipment was also new to NBG horticultural staff and so training was provided here too.

Stems were shortened to 15 cm, and the strongest and healthiest portions with some semi-ripe firm material at the base selected. Bundles of cuttings were fully immersed for 5 seconds in a solution of rooting hormone and water at the recommended concentration. They were then inserted into ground soil which had been dug and thoroughly weeded. Coconut fibre (referred to as 'cocopeat') and sand were dug into the soil beds to improve the consistency of the substrate, improve water retention, ensure adequate drainage and reduce the weed seed bank. Cuttings were inserted in separate rows for each collection number, with labels demarcating each collection (Fig. 11). Plastic cloches fastened over wire hoops were placed over the cuttings to retain moisture, protect the cuttings from excessive air flow and prevent disturbance. A piece of shade material was fastened over the plastic cover to reduce the effects of strong sunlight and maintain humidity under the cloches. A thermometer was placed in each cloche to monitor temperature ranges, and readings were noted daily.

Cuttings were checked every day and watered when necessary to maintain high

[6] https://rhizopon.com/en/products and https://rhizopon.com/en/rooting-guide?plant=5324&letter=T



Fig. 9 Cuttings were taken on arrival at NBG. Photo: Mark Watson.

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Fig. 10 Cuttings were inserted after soaking in IBA solution. Photo: Kate Hughes.

humidity. Training was given on aftercare, monitoring and record keeping. As cuttings were confirmed to have rooted, they were potted up into bags. The FTP collection number was kept with each collection.

Seed propagation

Seed was cleaned and the fleshy arils removed. Half the seed was sown immediately in trays of damp sand labelled with the appropriate collection number and placed on benches in open-ended polytunnels in the nursery at NBG. The other half was put in labelled containers of damp sand and into a fridge at 4 °C for some months.

Seed collected in 2018 was sown immediately on trays of sieved soil mixed with clean river sand (Fig. 12). As seedlings were pricked out the FTP collection number was kept with the plants.

Conservation assessments

A Red List assessment training workshop was conducted in May 2018. The purpose of the workshop was to build capacity and knowledge in the assessment process, and to gather data needed to undertake Red List assessments for the three species of *Taxus* in Nepal. Prior to the workshop, occurrence data were digitised for all three species using herbarium collections held at KATH, Royal Botanic Garden Edinburgh (E), RBG Kew (K), Natural History Museum, London (BM) and Tokyo University, Japan (TI). Added to this were field records collected by RBGE and Nepalese botanists and recorded on the Flora of Nepal database,[7] including the collections made during the FTP fieldwork. All 310 occurrence records were checked for accuracy in georeferencing and identification, and used to generate the distribution maps necessary for undertaking Red List assessments. These assessments were initially made in groups, refined during a feedback and knowledge-sharing session, and a consensus reached (Watson, 2018) (Fig. 13).

The conservation assessment workshops covered the following activities:

1. Introduction to the IUCN Red List, history and development, national and global assessments
2. Introduction to IUCN Red Listing categories and criteria used in assessments
3. Practical experience in carrying out Red List assessments using case studies from Arabia

[7] See the Flora of Nepal website at www.floraofnepal.org/data/specimens

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Fig. 11 Cloches were made and covered with shading material. Photo: Kate Hughes.

4. Practical experience in carrying out Red List assessments using case studies from Nepal
5. Formal National Red List Assessments for *Taxus contorta*, *T. mairei* and *T. wallichiana*

Training visit for Nepalese horticulturists

October 2019 saw the arrival in the UK of Kamal Nepali and Chandrakala Thakur, horticulturists and researchers at NBG and Brindavan Botanic Garden respectively. NBG and Brindavan are two of Nepal's twelve botanic gardens, run by the DPR. Kamal and Chandrakala spent six weeks in the UK, gaining experience of cultivating and managing plants in public gardens for display and conservation and working alongside the horticultural teams in RBGE's four Gardens. Activities



Fig. 12 Seed was cleaned and sown in trays. Photo: Kate Hughes.

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Fig. 13 Conservation assessment workshop. Photo: Mark Watson.

included maintaining the collections and records at Benmore Botanic Garden and Logan Botanic Garden, and in both the indoor collections under glass and the outdoor collections at the Edinburgh Garden. They used map drawing software to create a map for NBG onto which records could be added on their return to Nepal. Particular foci of their activities were record keeping and learning how to propagate ferns from spores (Hughes, 2021) (Fig. 14).

Results

Field collections

Four populations of *Taxus contorta* were surveyed in Manang District, and two populations of *T. mairei* were visited in Makawanpur District.

The quantity and maturity of seed available varied greatly among populations. Seed was more abundant in the western populations which were visited in 2018. Collections were gathered from twelve trees of *Taxus contorta* and three of *T. mairei* in 2017. Collections were gathered from 17 trees of *T. contorta* in 2018.

The propagation collections made on both field trips can be summarised as follows:

- 1 collection of seed of *Taxus mairei*
- 6 collections of seed of *T. contorta*
- 2 collections of cuttings of *T. mairei*
- 22 collections of cuttings of *T. contorta*

One year after collection many of the seeds sown had germinated, with a marked increase in the number of germinations 12 months after sowing (Fig. 15). At the time of writing, in 2022, of the 111 seeds that germinated, 101 are alive and growing well into young trees. Of the 114 cuttings that rooted, 88 have gone on to establish in pots (Fig. 16). As of 2022, therefore, NBG has 189

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Fig. 14 Patching off germinated fern spores. Photo: Kate Hughes.

plants in its conservation collection of *Taxus* species.

Conservation assessments

A group of DPR staff and staff from Nepal Academy of Science and Technology (NAST) with capacity in the Red List assessment process was established in the workshop. These staff are now ready to participate in further training to be accredited as IUCN Red List Assessors.

The IUCN website lists the West Himalayan yew (*Taxus contorta*) as Endangered (A2acd, IUCN Red List) (Thomas, 2011) with populations decreasing in the wild.[8] The East Himalayan yew (*T. wallichiana*) is also listed as Endangered (Thomas & Farjon, 2011). *T. mairei* is listed as Vulnerable – although its occurrence in Nepal is not given (Yang *et al.*, 2013). Participants in the workshop used their newly developed skills in the Red List assessment process to propose National Red List Assessments for these three species in Nepal. IUCN Red Lists were used as a starting point, along with newly produced distribution maps and the personal accounts of participants derived from observations made during fieldwork in Nepal over the last 40+ years (Poudel *et al.*, 2012; Bhatt *et al.*, 2017; Watson, 2018). Table 1 shows the assessments conducted as part of the Franklinia Taxus Project.

Discussion

NBG has a fully documented living collection of *Taxus mairei* and *T. contorta*. In 2022 some of the vegetatively propagated plants of

[8] www.iucnredlist.org



Fig. 15 Germination took 12–24 months. Photo: Kamal Nepali.



Fig. 16 Seedlings of *Taxus contorta* have established in polytubes. Photo: Dipak Lamichhane.

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Table 1 The new National Red List Assessments for the three *Taxus* species.

Species	National Red List Assessment
Taxus contorta	New National Assessment: ENDANGERED A2cd
Taxus mairei	New National Assessment: CRITICALLY ENDANGERED B2ab (iii, iv, v)
Taxus wallichiana	New National Assessment: ENDANGERED A2acd

T. contorta reached an appropriate size and were planted out at NBG in two different areas: the Biodiversity Education Garden and the Taxonomy Family Garden. They will be used for education and as a resource for further research into the species. Where the prevailing climate aligns with the needs of the species they can be distributed to other botanic gardens. For example, Mountain Botanic Garden at Daman is located near to the dwindling populations of *T. mairei* and the distribution of plants to that garden may be appropriate.

One of the outcomes of the Franklinia Taxus Project is the enhancement of horticultural capacity in DPR. There is increased capacity in propagation from semi-ripe cuttings and seed at NBG. Other DPR gardens also have enhanced horticultural capacity: Chandrakala Thakur has had success with experiments in the cultivation of ferns from spores at Brindavan Botanic Garden (Hughes, 2021). She germinated a small number of plants of *Diplazium esculentum* and *Tectaria coadunata* in June 2021. In 2022, she successfully germinated spores of the CITES-listed species *Cyathea spinulosa*, and young sporophytes are being brought on for the first time (C. Thakur, pers. comm.; Hughes, 2021) (Figs 17 & 18).



Fig. 17 Cultivation of ferns from spores, Brindavan Botanic Garden. Photo supplied by Chandrakala Thakur.



Fig. 18 *Cyathea spinulosa* grown from spores at Brindavan Botanic Garden. Photo: Chandrakala Thakur.

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Updated conservation assessments from the Red Listing workshops are valuable, but represent a cause for serious concern with regard to the continuance of the three yew species of Nepal. The knowledge shared at these workshops informs conservation programmes and is available through online databases (Thomas *et al*., 2019).

Conclusions

The Franklinia Taxus Project has generated invaluable collections and information on which to base future projects to safeguard the three yew species of Nepal. The living and preserved collections and data gathered are stored in Nepali institutions and are accessible to Nepali researchers to develop future programmes. The expansion of cultivation techniques in Nepalese botanic gardens is key to the development of *ex situ* conservation programmes. The living collections of *Taxus mairei* and *T. contorta* at NBG and spore-grown ferns at Brindavan Botanic Garden are an excellent resource for future conservation programmes.

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