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Tamar Kurdadze collecting seeds of yew.

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Collecting rare, endangered and protected woody plants in Georgia

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Almost 10% of Georgia's flora is made up of around 400 tree and shrub species. Unfortunately, many of the woody species face a wide array of threats, and Georgian conservationists are searching for effective policy mechanisms to ensure sustainability of these endangered trees and shrubs. The Global Tree Seed Bank Project, funded by the Garfield Weston Foundation, is an important step in preserving the unique genetic resources of Georgian woody flora.

The Global Tree Seed Bank Project aims to secure the future of more than 3,000 of the world's rarest, most threatened and useful trees. The Institute of Botany of Ilia State University (IoB) has been involved in this project since 2015. With partners from the Royal Botanic Gardens, Kew and the National Botanical Garden of Georgia (NBGG), the IoB team will be responsible for collecting seeds and herbarium material until 2019 of 33 Georgian woody plant species known as relic, endemic, rare, endangered and legally protected taxa.

In the summer and autumn of 2015, the IoB team collected 11 woody species – five are included in the Georgian Red List, two are Georgian endemics and one is endemic only to the Caucasus region. A list of potential locations of target species populations was compiled prior to field work, based on data extracted from herbarium labels, available

literature sources and advice provided by colleagues from other research institutions. Several local nature enthusiasts and amateur botanists were also consulted, to identify access routes to the location of some species growing in remote areas without access for vehicles.

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Georgia is a country, with a varied range of habitat and vegetation types – from lush seaside forests to alpine scrub. As a result, the field team had to travel to different parts of the country to collect seeds from target species, ranging from imposing deciduous giants like the 35 metre tall Caucasian elm (*Zelkova carpinifolia*), to dwarf evergreen shrubs, like the Caucasian rhododendron (*Rhododendron caucasicum*), reaching only 1.5 metres. The team visited six regions, including coastal areas with a near subtropical climate in the south-west, and dry territories in the south.

Unfortunately, the majority of woody species in Georgia are threatened by anthropogenic impacts like tree felling, grazing, trampling, road construction, tourism, and the overexploitation of economically valuable plants. Some species, like the yew, were already on the verge of extinction during the middle ages. In the twelfth and thirteenth centuries, the legendary Georgian Queen Tamar forbade the cutting of yew trees in order to preserve the species within the forests of Georgia. Since then, the Georgian name for this plant has been *Utkhovari* – 'do not ask me for'.

Not all the target species and their populations were easy to find. Two blackberry species were well hidden in the relic deciduous woodlands of Lagodekhi National Park in East Georgia and it took an experienced taxonomist's eye to spot the plants among other shrubs in the undergrowth, and collect their cylindrical fruits. Limited numbers of other species are widely scattered across woodlands, like the Georgian Red List species hop-hornbeam. Only 19 randomly dispersed trees were found in the mixed woodland near the popular resort of Borjomi in South Georgia. Happily, efforts to locate hop-hornbeam trees were not in vain, and around 3,000 hop-like seeds of this threatened species were collected

Not all tree species in Georgia have been as long-endangered as the yew. One target species, the Caucasian elm, was once widespread in West Georgia, and known as the most common species in that area as recently as the early 1900s. The Caucasian elm is known for its durable timber, widely used to build boats. The Latin name of this plant, Zelkova, appropriately stems from Georgian dzeli kva, which means 'beam stone'. Its popularity in construction meant it was over-exploited and became fairly rare in Georgia, and only small patchy populations remain. To prevent further decline, the Caucasian elm was included in the Georgian Red list and some 1,000 seeds of this species were collected by the IoB team in Imereti region in West Georgia last year.

Another important component of the Global Tree Seed Bank Project is documenting the biotic and abiotic characteristics of the target woody plant population, to build our knowledge of these endangered and potentially useful species. Recording the locations of tree and shrub populations with a hand-held GPS contributes to our understanding of the distribution of species. Identifying current threats will help us develop conservation strategies and ensure the sustainability of wild populations into the future.

IoB's long-term partner institution – NBGG – is responsible for processing and



loB team member, Marina Eristavi collecting seeds of Caucasian elm in a roadside woodland patch in Terjola municipality, Imereti region of West Georgia.

depositing the seeds into the Caucasus Regional Seed Bank, maintained by their Department of Plant Conservation. The collected herbarium material is deposited in the loB's National Herbarium of Georgia, and the duplicates of both the seeds and herbarium materials are shipped to RBG Kew for storage at the UK's Millennium Seed Bank. The long-term partnership between the UK and Georgian research institutions is a legacy from the first phase of the MSBP. More than 1,000 species have now been safeguarded, and together we are continuing to target more species for collection in the coming years.



loB team member, Tamar Kurdadze collecting seeds of Georgian Red List species yew in the understorey of deciduous woodland on the slope of Mt. Zedazeni to the NW of the Georgian capital Tbilisi.



A message from Colin Clubbe

(Head of the Conservation Science Department, RBG Kew)



As we go to press, Kew has just published the first *State of the World's Plants* report (www.stateoftheworldsplants.com). This will be an annual horizon-scan of the global status of vascular plants.

The three sections cover what we know about plant diversity, the urgent threats that plants are facing globally, and a look at how key policies are working. We are very grateful to the Sfumato Foundation for the generous funding that will enable this review until the close of the UN Decade on Biodiversity in 2020 (www.cbd.int/2011 2020). This will be an important year, when the international community reviews the status of global biodiversity, and how far the Aichi Targets have been met. Our annual *State of the World's Plants* will be a key resource for this review process. Trees are iconic plants and a vital component of plant diversity – underpinning many ecosystems, the services they provide and the livelihoods they support. It is no surprise that trees feature strongly in the report, from newly identified species and the Global Conservation Assessment of all Trees, to the ongoing loss of huge tracts of forests to other land uses.

This issue of Samara richly illustrates many of Kew's projects with its national and international partners, with many examples of how these are feeding into international commitments and contributing to a better outlook for trees and the biodiversity they support. Without this vital conservation work, the world would be in a much worse state and people the poorer for it.

Thank you all for being part of the MSBP.

A message from Jonas Mueller

(Senior Research Leader, Seed Conservation, RBG Kew)

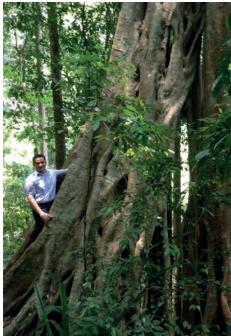


I am just coming back from my lunch break, which I spent walking in the grounds of Wakehurst Place. These are the days when many visit Wakehurst to see our magnolia trees in full flower. Further down into the valley, towards Westwood Lake, is our pinetum. Those mighty conifers, many of which were planted in the nineteenth century, remind me of the role that tree species play on our planet, in providing ecosystem services or simply giving aesthetic pleasure to us humans.

Every edition of the UN Food and Agriculture Organization's *State of the World's Forests* cites ever-growing recognition that forests and their use lie at the centre of any serious discussion about a sustainable future for our planet – providing forest products and ecosystem services essential for our wellbeing. The last edition highlighted the socioeconomic benefits of forests, and even how they contribute to food security.

Nobody knows exactly, but reliable estimates suggest there are 50,000 or more tree species in the world - most of them in the wet tropics. It is no surprise that many across the Millennium Seed Bank Partnership have made tree seeds the focus of their work and are involved in major projects about seed biology, physiology and the practical conservation of trees and their seeds. This edition of Samara highlights some of these projects. I would like to draw attention to a couple of them: The Garfield Weston Global Tree Seed Bank Project, whose collecting and research activities in 14 major forest regions worldwide are now in full swing - you will find examples from the Caucasus and the Americas in this issue, as well as short articles on ongoing scientific work looking into the storage conditions of recalcitrant seeds. And the UK National Tree Seed Project, focusing on our native British tree flora. The need to use the right sampling strategy in the field, to capture as much diversity as possible, is highlighted in a separate article.

We already store high quality seeds of more than 4000 tree species here at the Millennium Seed Bank. Partner seed banks store additional species and the number of tree species banked continues to increase steadily. I would like to hear more about your own interest in and work with tree seeds!



Jonas Mueller providing scale with Ficus altissima.

The Global Tree Seed Bank Project

SHARON BALDING (Seed Conservation Projects Officer, RBG Kew)

aunched in 2014 with generous funding from the Garfield Weston Foundation, the Global Tree Seed Bank Project is going from strength to strength. New and existing partners have been targeting the collection of seeds from rare, threatened and useful tree species, as well as developing propagation protocols and restoration programmes for some key species. A research programme, including four post-doctoral research projects, has also been established to focus on important questions for tree conservation.

The project aims to conserve 3,000 of the rarest, most threatened and useful trees, and to undertake research to improve our knowledge of tree conservation. Funded in two phases, the first targets regions of the world where trees are particularly threatened: Madagascar, Africa, the Dominican Republic, Mexico and Australia. In addition to collecting seeds, the research will improve our knowledge of the seed biology, germination and propagation of key species, to develop successful reintroduction programmes. Notable species collected include: Tarenna hutchinsonii, a Rubiaceae species from coastal Guinea. listed by the IUCN as Critically Endangered;

'The project aims to conserve 3,000 of the rarest, most threatened and useful trees, and to undertake research to improve our knowledge of tree conservation.'

Pseudophoenix ekmanii, a palm from the Dominican Republic, also Critically Endangered; and *Guaiacum sanctum*, a Caribbean species in the Zygophyllaceae family, listed as Endangered.

The second phase will accelerate and expand the project by broadening the geographic range and filling notable gaps in our knowldege of tree conservation. Further partners in Africa, South East Asia, Europe, the Caucasus, the Caribbean and the South Pacific have joined over the last two years. Four post-doctoral researchers have now been appointed to work on major areas of tree conservation. These include:

- developing generic protocols for banking recalcitrant (desiccation sensitive) seeds.
- analysing the functional seed traits of palms, to improve collection strategy and storage protocols.
- compiling DNA sequence data for all genera of South East Asian trees, to explore links between conservation and evolution. This will also be useful for authenticating and regulating important timber species.
- studying the evolution and diversification of desiccation sensitivity in the Asia Pacific region, using palms as a model family.

It is a mark of the progress made so far that a number of the articles in this edition of Samara describe some of the research and projects of the Global Tree Seed Bank, including seed collecting in Georgia, reforestation in Mesoamerica, and research into important recalcitrant species in Europe.

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MSB's Roberta Hope providing field-based training in Thailand as part of the Global Tree Seed Bank Project.



A participant, Thaveechok Jumruschay, on the training course in Thailand using sieves.

Supporting reforestation through conserving seeds of useful native trees in Mesoamerica and the Caribbean

EFISIO MATTANA, MICHAEL WAY, HUGH W. PRITCHARD, TIZIANA ULIAN (RBG Kew) OSWALDO TÉLLEZ-VALDÉS (Universidad Nacional Autónoma de México, F.E.S. Iztacala, Mexico City, Mexico) FRANCISCO JIMÉNEZ RODRÍGUEZ (Jardín Botánico Nacional 'Dr. Rafael Ma. Moscoso', Santo Domingo, Dominican Republic)

he Global Tree Seed Bank Project is one of Kew's major science-based plant conservation programmes. Funded by the Garfield Weston Foundation, it aims to secure long-term storage of seeds from at least 3,000 tree species from across the world.

'… one of Kew's major science-based plant conservation programmes'

In Latin America, the programme started in 2015 through two projects focused on useful trees. These are managed by the Natural Capital and Plant Health Department of RBG Kew in collaboration with the F.E.S. Iztacala, Universidad Nacional Autónoma de México (UNAM) and the Jardín Botánico Nacional Dr. Rafael Ma. Moscoso (JBN) in the Dominican Republic. The 'Science-Based Conservation of tree species in Mexico' project is integrating *in situ* and *ex situ* conservation of endemic and useful plants, protected through listing in the National Official Laws, by the IUCN and CITES and which are important for the livelihoods of rural communities.

The main objectives are:

- producing a list of tree species of Mexico, with information on distribution and conservation status along with their associated uses and a map of tree species biodiversity hotspots
- long-term conservation of seeds of 200 tree species
- seed germination and plant propagation protocols for at least 25 priority useful tree species
- a predictive model on seed desiccation tolerance.

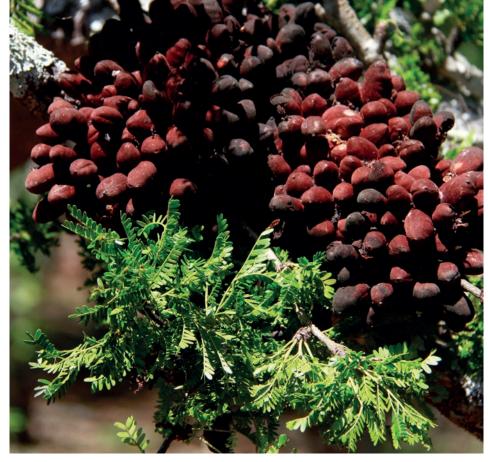
In the Dominican Republic, the 'Saving threatened forests of Hispaniola' project will protect the forest diversity in Hispaniola by:

- conserving seeds of at least 200 tree species
- screening seeds of at least 100 tree species for desiccation tolerance
- propagating 25 priority tree species, to support reforestation.

These four-year projects will support reforestation through *ex situ* seed conservation and researching useful wild trees.



The FES Iztacala Team collecting tree seeds in the Chamela-Cuixmala biosphere reserve (Jalisco, Mexico).



Arcoa gonavensis Urban (Tamarindo cimarrón), endemic of Hispaniola.



The UK National Tree Seed Project

CLARE TRIVEDI (UK Conservation Partnerships Co-ordinator, RBG Kew) and SIMON KALLOW (UK National Tree Seed Project Officer, RGB Kew)



Tagged mother tree.

he UK has one of the lowest rates of woodland cover in Europe, just 13% of its land cover. Though much is publicly accessible, most of this cover is privately owned and there is relatively little state intervention, compared to other countries, in the management of this precious resource. Nevertheless, UK woodlands are highly valued as places for recreation and for the ecosystem goods and services they provide. The need to expand our woodland cover has been recognised, along with the value of using native species to do so. However, several challenges lie in the path of meeting these aspirations. Our trees and woodlands are at high risk from pests and diseases, and foresters and conservationists are struggling to predict and plan for the uncertain impacts of climate change.

In this context, RBG Kew launched the UK National Tree Seed Project (UKNTSP) in 2013 as part of the MSBP. This ambitious project aims to make 'geneticallyrepresentative' collections of the UK native woody flora. These need to be made and stored in ways that will make them as useful as possible for studies and activities, seeking to overcome the challenges to UK woodlands. To address this need, the UKNTSP is stretching MSBP standard practice to new limits.

How to make 'genetically representative' collections?

A problem we share with most countries is that we don't know enough about the population genetics of UK trees and shrubs. Therefore, it can be difficult to be sure how to achieve collections which are genetically representative of the national flora. 'These collections need to be made and stored in ways that will make them as useful as possible for studies and activities which are seeking to overcome the challenges to UK woodlands.'

Fortunately, the UK Forestry Commission (our state forestry agency) had already divided Great Britain into 24 biogeographic areas, called 'native seed zones', for the purposes of seed collection and supply. This gives us a good proxy for likely areas of genetic diversity. Great Britain also has good species distribution records, though it can be hard to tell whether records are of native populations or planted ones. We mapped the distribution records across the country and overlaid it with our native seed zones map -thus developing a database of target collections. We hope to collect from every seed zone in which these species naturally occur. Where species are found both above and below 300m altitude, we will attempt to make a collection from both altitude zones too.

Developing a list of target collections is only the start. It is equally important that the collections made adequately represent the populations (or sub-populations) they come from. This is an issue we struggled with, as standard seed-banking advice suggests collecting from at least 50 individuals, well-spaced through the population. Unfortunately, UK woodland is highly fragmented, often meaning this many individuals are simply not found in one woodland. In addition, tree seed collecting is time consuming. At the individual tree level it is vital that seed is collected right across the canopy, to catch the progeny from different fathers. We found a team of 3–5 people can only collect from 10–20 trees per day. Collectors are having to make informed but pragmatic decisions to maximise the genetic diversity captured – often collecting from dispersed sites across the seed zone and/or collecting from a reduced number of individuals.

Having worked so hard to capture the maximum diversity in our sample, we don't want to lose access to this once seeds go into storage. We therefore tag and georeference individual mother trees and store their seed separately. This allows current and future users of the collections to study traits such as disease resistance for maternal lines, as well as at the population level.

We are also working with geneticists to improve this sampling strategy. Desk studies for all target species have revealed what is known about their population genetics, and provided species-specific guidance for their sampling. We are also working with Sean Hoban (see article pp. 8–9) to model the effectiveness of our collections in capturing the genetic diversity available in the wild. An additional research component to the project, to better understand germination and storage of our UK trees and shrubs, is being carried out by germination specialists and students.

The last three years have been a time of intense learning for us. We are keen to share experiences with MSBP colleagues around the world who are either embarking on a national tree seed collecting project for the first time, or who have years of experience behind them. Please do get in touch.

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Finding the woods and the trees

BEDE WEST (UK National Tree Seed Project Fieldwork Officer, RBG Kew)

here are 3.15 million hectares of woodland in the UK (Forestry Commission 2015), but an average size of only 9 hectares (Forestry Commission 2011) means our woods are highly fragmented and very difficult to survey. Yet the UKNTSP team at Kew must do just this to locate and collect target species not covered by our partners. So how do we find the woods and the trees?

'So how do we find the woods and the trees?'

The simplest method for finding target species is approaching large public and private landowners who manage diverse woodlands. This allows us to collect as many target species as are present from across the landowner's sites, and is helpful when the list of target species is diverse and long. When searching for specific target species, however, we recommend using public biological records with follow-up enquiries and site visits to confirm population suitability.

Once we have located a suitable target population, we collect seed using one or more of the four main methods best suited to collecting seed from wild trees. To ensure the parent is known, we never collect seed from the ground. In order of their efficiency, the methods are:

- direct picking the simplest method of collecting tree seed. However, this is only practical when branches with seed can be reached. Picking can be done by hand or with tools like secateurs and berry pickers.
- pole pruning seeding branches (pictured) is the preferred method for the UKNTSP, as this is fast and easy. Pole pruner heads and poles that extend up to 10m (sometimes more), are easily purchased in aluminium, fibre glass or carbon fibre, or can be made fairly easily from wood or metal.
- shaking and pulling branches, with throw lines or poles – using a spread out tarpaulin to collect any falling seed is a simple method which can reach as high as you can get a throw line or pole. However, this relies on seeds detaching easily from the tree and not blowing away, so care must be taken when placing the tarpaulin and during shaking for some wind dispersed species.

 climbing – rarely practical, because of the time and effort required compared to other methods, and not worth considering unless tree canopy base or seeding branches are over 20m up. It takes 10–30 minutes to climb up a UK tree safely, depending on the size of the tree and the competence of the climber. The variability of terrain and trees mean it is often impractical to use assistive methods like a throw line launcher, ladder or elevating platform to get a safe access rope anchored in the tree.

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> Pole pruning to collect UK tree species.

The John Muir Trust

MIKE DANIELS (Head of Land Management, John Muir Trust)



Hawthorn seed collection on the Isle of Skye.

he John Muir Trust is a conservation charity dedicated to protecting and enhancing wild places. We manage some of the finest wild places in Scotland, including the summit of Ben Nevis and part of the Cuillin on Skye. Working with RBG Kew's UKNTSP fits in with our science based approach to land management and our desire to protect and enhance wild land ecosystems including native woodlands.

Collecting seeds on our properties is particularly challenging, due to the nature of the land and the woodlands we manage. Wild land is, by definition, relatively remote and inaccessible, so all of our collections have to be made on foot and sometimes after a good hike!

We had a successful and varied first year in the field, and we plan to continue our collections in the autumn. We hope that the project and the information collected will help raise the profile of our threatened native trees, as well as providing useful genetic information. Most of all the John Muir Trust is pleased to do its bit.

Gwent Wildlife Trust

ANDY KARRAN (Wildlife Sites Officer, Gwent Wildlife Trust)

went Wildlife Trust is the leading wildlife conservation charity in Gwent (Wales, UK), working for the wildlife and people of this rich and varied landscape. Four UK seed source zones fall within Gwent, and we will be collecting tree seeds from three of these until 2017.

Gwent is blessed with many great habitats, including plentiful woodlands. These are a mix of conifer plantations, secondary broadleaved woodland and ancient semi-natural woodland – the Wye Valley woodlands in the east, and the highest naturally occurring beech (*Fagus sylvatica*) woodland in the UK on our Silent Valley reserve in the west, are justifiably famous.



Crataegus monogyna - ready to be processed.

Seed collection for the UKNTSP is a great experience for both staff and volunteers, and got us out in lovely woodlands on some balmy autumn days as the leaves were changing colour. Getting the seeds from the trees was challenging but fun, using throwlines, telescopic pruners and shaking branches. Collecting the seeds and fruit was only part of the fun; we then had to extract the seeds. We are looking forward to this year's collections.

Improving the sampling of seeds for conservation

DR SEAN HOBAN (Tree Conservation Biologist, The Morton Arboretum) and MICHAEL WAY (Conservation Partnership Co-ordinator, RBG Kew)

proximately one-fifth of all plants globally are threatened (Brummitt et al. 2015). While some can be maintained *in situ* through sustained conservation measures, others must be preserved in *ex situ* seed banks or botanic gardens due to imminent threats to wild populations. Seed banks also provide diverse seed to researchers, plant breeders (horticulture, agriculture and forestry), and restoration managers.

'Seed banks are most useful when they represent as much of the diversity of their wild populations as possible'

Seed banks are most useful when they represent as much of the diversity of their wild populations as possible. This captures the most traits and genes for improved agriculture, or the most adaptive capacity for a species to evolve under environmental change. Of course, limited time, funds, and space mean we can't sample seed from every living plant. Seed collectors must strategically plan how much seed to collect from which populations, in order to maximize the variation preserved, while avoiding damage to the wild population and keeping within their budgets.

This article reviews past and future seed collecting strategies, to identify improvements to seed sampling for conservation.

How good are the existing sampling guidelines?

There are various methods for prioritizing which species we would conserve first, including ranking them by phylogenetic distinctiveness, endemism, conservation status, and their importance to ecosystems or to humans. The methods we use to prioritize our sampling within each species - which populations and how many, for example - are less well articulated. Nearly all collectors have referred to a universal 'rule of thumb' in deciding their sampling strategy: 50 samples per population from 50 populations, or as many as can be reached (Brown & Marshall 1995). About two-thirds of leading seed collecting organizations use this protocol as the starting point



Collecting seed from Cercocarpus montanus, Alderleaf mountain mahogany.

a Number of plants sampled

The authors have analysed data from over 5,000 North American collections for the MSB since the 1960s. The results show that about 75% of all tree seed collections are from fewer than the Brown & Marshall recommended 50 plants, with a median of 30 plants (Figure 1a and b) which is actually the guideline for known outcrossing species. However collectors did compensate by generally sampling from more seeds per plant (Figure 1c, linear regression of log-transformed variables). Though threshold seed numbers were often achieved, some collections may be under-representing the diversity of these species due to 'diminishing returns' of sampling on a given tree.

b Number of plants sampled

in each collection all plant life forms in each collection only trees more seed per plant and sampling from more plants under of plants sampled not plants sampled n

c Trade off between sampling

Figure 1: Sampling in practice: analysis of Millennium Seed Bank collections from North America.

for sampling (Table 1 in Hoban & Strand 2015), including Seeds of Success and the Millennium Seed Bank Partnership.

However, plants differ widely in life form, distribution, abundance, and mating system, and theoretical and empirical research over the past decades has demonstrated that a species' genes and traits are distributed very differently depending on that species' characteristics (Dick et al. 2008). If each species' genetic diversity is distributed differently on the landscape, and differently among individuals and their seeds, perhaps we should tailor our sampling for different species. Indeed, members of the Center for Plant Conservation (CPC) have recommended sampling according to species characteristics and life history (Guerrant et al. 2004) - sampling more individuals in species with high rates of selfpollination, due to lower diversity in a given seedset, for example. Such suggestions are helpful general advice, but they don't enable collectors to determine 'how much' for a target species – '50 plants, plus additional ones' will be interpreted in different ways by different people. Formal and quantitative guidance may be needed.

To meet the anticipated needs of users, collectors are also often seeking threshold seed numbers in their collection (typically 10,000 seeds), while sampling from representative plants across the population. Collectors must make decisions about tradeoffs while in the field, as circumstances change. Trees, for example, are much more time-consuming to access and sample from than grasses, but one tree may yield thousands of seeds. In practice, do collectors sample more seeds from fewer maternal trees? (see Figure 1).

Does one size fit all?

Research by Hoban and collaborators shows that various plant characteristics can guide a nuanced and optimal sampling protocol. In short, one simple protocol does not apply well to all species. For example, Hoban & Strand (2015) have developed an approach to calculate precisely what amount of genetic variation might be captured in a sampling protocol (number of maternal plants and number of seeds per plant) for a 'typical' species that exhibits high selfpollination compared to low self-pollination, and that exhibits extended seed dispersal compared to reduced seed dispersal. These data allow us to see, as predicted by Guerrant et al. (2004) and others, that self-pollinating species need more intensive sampling (more plants per population) and fragmented species need more extensive sampling (more populations). The innovative aspect of this work is that researchers can estimate 'how much', with a number to aim

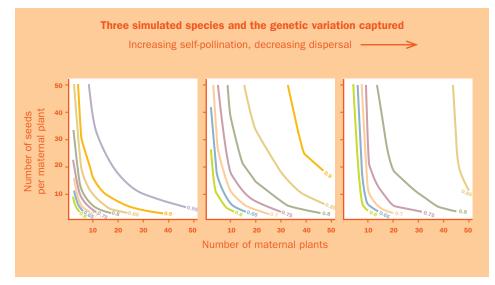


Figure 2: Plots showing change in the amount of genetic diversity (coloured lines) captured in a given number of seeds and plants, depending on pollination and dispersal traits.¹

for. For example, their data suggest that to capture 90% of the diversity of a lowdispersal, highly self-pollinating species, collectors would need to sample from sufficient additional plants and seeds per plant, in order to achieve an overall seedlot size five times as large (Figure 2).

Conclusions

As early as 1991, the CPC suggested that collectors should review information about their target species with a desk study prior to fieldwork. The recent work by Hoban & Strand confirms and provides more details about what information is needed at that stage: self-pollination, seed dispersal, degree of fragmentation (see also Hoban & Schlarbaum, 2014), and annual/perennial life-span are all important. These traits influence how much variation is in a seed sample, and whether 50 samples have as much variation as we think.

Of course, this work continues and it is expected that other factors will also be important. While seed dispersal is known to strongly influence spatial genetic structuring, seed morphology is complex, and dispersal is hard to estimate. It may be possible to clarify easier characteristics either before collections or during fieldwork. Two examples are clonality and recent population history. Recently founded or highly clonal species may need only a few samples from those sites, because low diversity may be present to begin with. Ancient population history is likely to be important too: highlatitude populations (which migrated farthest since the last Ice Age) could be best sampled differently from mid-latitude ones. The eventual use of the seed (for either breeding/conservation/restoration) should, of course, be another consideration.

Genetic studies, desk studies and further use of the simulation approach of Hoban and collaborators will help refine these ideas. Collection programmes that provide tissue for genetic diversity analysis will also be able to adjust their sampling according to the actual diversity encountered. In short, we know much about the basic science of the partitioning of genetic variation, and a new wave of research is helping to translate this to sampling guidelines. Through a combination of desk studies and field observations, we believe that collectors can improve the cost effectiveness and the sampling of seed for conservation.

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Growing farm-based green enterprises: the work of the Citi Entrepreneurs Project in Kenya

HUMPHREY GAYA, PRISCA MAKENA (KEFRI Citi Project Office) and TIM PEARCE (Conservation Partnerships Coordinator for Africa, RBG Kew)



Jackob Njiru's Citi nursery in Mbeere.

long-standing concern of many Kenyan agencies is that there are insufficient sources of high quality tree planting material of indigenous species to fulfill the national tree cover aspirations in the Government's Vision 2020 targets.

As a result, this project set out to establish how a measurable increase in the availability of indigenous tree seedlings could be achieved, while also showing a sustainable increase in farm-based income, by growing and selling indigenous trees and their seeds.

In 2012, we were awarded three years of funding from the Citi Foundation. Operating in Tharaka, Mbeere, Siaya and Nyamira Districts, the project has been managed by the Kenya Forestry Research Institute (KEFRI), with input from Farm Concern International (FCI) and the Mount Kenya University in Thika.

At the heart of the project lies the development of a training programme aimed directly at providing a suitable level of both nursery and business skills for our farming

Jackob has become a model for farm-based enterprise in his Mbeere region.'

students. This has been complemented by a series of market reports on the actual and potential demand for seedlings of indigenous trees.

By the end of the project period we have trained some 115 farmers, and more than 40% of them are still actively involved in tree seedling production and supply. They are operating independently as profitable businesses with no external financial support from Government or the project. At least 15 of our trainees cite tree seedling production and supply as a 'new' and 'significant' contribution to their farmbased income.

One of the successful entrepreneurs is Jackob Njiru who hails from Mbeere. When the Citi project selected him as a trainee, Jackob was already interested in growing indigenous trees and had a small nursery holding a stock of about 2,000 seedlings. Many of these he planted on his own land or gave to his friends and neighbours.

Since graduating as a Citi Enterpreneur, Jackob has become a model for farm-based enterprise in his Mbeere region. He now runs a significant tree nursery, containing 50,000– 100,000 predominantly indigenous tree seedlings at any given time, depending on his orders. He has excelled in marketing, and now sells his tree seedlings in Embu County as well as in the neighbouring counties of Kitui and Meru; a feat he readily admits he could not imagine before the training.

He now runs occasional seedling production training courses using his nursery as a model for demonstration, even for the local Kenya Forest Service (KFS) authorities. Jackob now keeps impeccable records of his activities including input, expenses and income reflecting the progress he makes every day. He has employed five more people and has embarked on an expansion programme, opening satellite nurseries in the neighboring County of Kitui in which he has posted two employees.

Jackob has grown his customer base and now supplies to individuals, local and national institutions, NGOs, churches, county governments and even international organisations where he has been enlisted as a supplier in the competitive bidding processes. He is now a registered seed supplier for KEFRI, a registered seedling supplier for KFS and is also a recognised supplier of both indigenous and exotic tree seedlings in the county government system. He has now branched out into promoting and training in the use of improved charcoal production kilns; a partnership he has forged with UNDP.

Jackob's story is just one example emerging from the Citi Entrepreneurs Project. With his new house and children now at boarding school, there is no doubt his green enterprise has helped Jackob in life. But can this model of success be expanded countrywide and truly provide the high quality materials needed to reach Kenya's ambitious planting target? The answer is yes, we think it can. We are currently developing the next phase of the programme, with high hopes of it meeting future ambitions.

Rare monkey puzzles at Wakehurst

JO WENHAM (Plant Propagation and Conservation Manager, RBG Kew)

n evergreen conifer native to Chile and Argentina, monkey puzzle (Araucaria araucana) is one of 19 species in the genus Araucaria. Fossilised specimens of 200 million year old Monkey puzzle trees have been found in rocks of the Jurassic Period. Although these ancient monkey puzzles are now a wellknown garden tree, few people are aware that they are under threat in the wild as a result of anthropogenic activity like illegal logging fires, and agricultural clearances in particular. The monkey puzzles found in Chile's Nahuelbuta National Park form the remaining genetic coastal stronghold, making this population even more vulnerable than the one found in the Andes.

Araucaria araucana cannot be stored in conventional seed banks, so seeds from this species are propagated for planting and will be conserved in the landscape at Wakehurst, RBG Kew's country estate. The plants were grown from seed collected by Plant Propagation and Conservation Manager Jo Wenham and fellow Wakehurst experts, MSBP partners at the Agricultural Research Institute of Chile (INIA), and the UK Forestry Commission. Jo's team travelled through leech-infested rainforests and a smoking volcano to hunt for rare and threatened plants. 'The experience has continued to weave through my professional and personal lives. The jaw-dropping landscape of Chile is astounding, but no more so than the pride that comes with planting seeds and then trees you have collected in the wild.'

A team of more than 25 horticultural staff and volunteers have already planted around 40 rare monkey puzzle trees, in a new Chilean area being created in Coates Wood at Wakehurst. More species from the expedition will be propagated and planted out over the next two years. Eventually, a landscape will be created that will allow visitors to experience something close to a walk up a Chilean mountain side. This is just the start of a project which will see many more seeds from Chilean threatened species being propagated and planted at Wakehurst.



Jo Wenham planting a monkey puzzle at Wakehurst.

Ed Ikin, new Head of Horticulture and Landscape, said 'Wakehurst is a place of wild landscapes, where the plants of farflung countries like Chile thrive in the rich soil and undulations of the High Weald. Our monkey puzzle grove will immerse our visitors in one of Chile's iconic landscapes, while delivering Kew's mission to conserve threatened plants worldwide'.

Tragacanth milk vetches of Armenia

DR. I. AREVSHATYAN and DR. A. NERSESYAN (Seed Bank of Armenian Flora – Institute of Botany of the Armenian National Academy of Sciences)

ragacanth Astragalus species (Fabaceae family), also known as tragacanth milk vetches, are densely foliose plants around 10–70 cm tall with multiple branches. Their leaves are paripinnate, with rigid, spiny axes that remain on branches when leaflets have fallen. The flowers are generally axillary, and are either sessile and gathered in condensed raceme inflorescence, or on the apices of flower stalks. The fruit is not exerted from the calyx and contains only one or two seeds. Tragacanth milk vetches produce gum in their pith and pith rays. This gum is valuable in various industries.

The tragacanth milk vetches are distributed across the ancient Mediterranean region, with the highest species diversity found in the eastern part of the area. Tragacanth *Astragalus* species occur mostly in steppe, semi-desert, and where phryganoid¹ plant communities are found. The species occupy open, dry habitats with thin skeletal soils, as well as over-grazed habitats. Some species, however, are found along the alpine belt,



Astragalus aureus.

growing alongside typical alpine plants. An example of these high-mountain species includes *A. lagopoides* Lam., which occurs in altitudes of 1,400–3,000 metres above sea level. Others include *A. aureus* Willd. (1,500–3,100 m.a.s.l.) and *A. microcephalus* Willd. (800–3,000 m.a.s.l.). Some species such as *A. uraniolimneus* Boiss. and *A. divaricatus* Bunge only grow in altitudes above 2,000 m.a.s.l. and prefer grassy, stony slopes. In Armenia, more than 120 *Astragalus* species can be found, of which 16 are tragacanth milk vetches. The species *Astragalus* microcephalis, *A. compactus* Lam., *A. aureus* and *A. lagopoides* are found frequently in Armenia. According to field observations, these species produce a good amount of seeds, with adequate dispersal rate and also regenerate well from seed. In contrast, the species *A. amblolepis* Hohen., *A. divaricatus, A. karabaghensis* Bunge, *A. oleifolius* DC., *A. longifolius* Lam., *A. basianicus* Boiss. et Hausskn., *A. meyeri* Boiss., *A. karjaginii* (Boriss.) Boriss., *A. sosnowskyi* Grossh., *A. uraniolimneus* and *A. szovitsii* Fisch. & C. A. Mey. are relatively rare in Armenia. Another rarely found species is *A. vedicus* Takht., which is an endemic species for the Southern Transcaucasia (Armenia and Nakhichevan).

Interestingly, morphological, taxonomical and geographical analysis of tragacanth milk vetches of Armenia found that they are more closely related to tragacanth Astragalus species found in Anatolia and Iran, than those found in the northern regions of the Caucasus.

A type of community that is dominated by dwarf xeromorphic shrubs

State of the World's Plants

EMMA WILLIAMS (Species Conservation Assessor, RBG Kew)

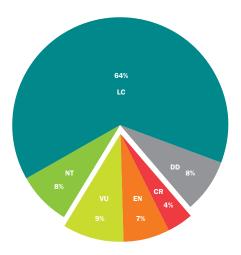
he State of the World's Plants report is one of nine outputs from our Science Strategy 2015–2020. The first report and symposium, in May 2016, provided a cutting edge view of the status of the plant kingdom. Kew scientists and partners combined their extensive knowledge and expertise to evaluate the current, global status of plants. Some of the questions explored included:

- What is our best estimate of how many plants are threatened with extinction?
- How many plant species are classified as invasive?
- How is climate change affecting plant species, populations and communities globally?

In addition to providing new evidence and perspectives on a range of key issues, the report helps identify strategic research and policy priorities for both the UK and overseas.

One of the key components of the report will be assessing the extinction risk of plant species. To quantitatively assess the level of risk, Kew will continue its work on the IUCN Sampled Red List Index (SRLI) for plants. The IUCN Red List records the extinction risk for some 80,000 organisms globally, using a rigorous set of criteria to evaluate the relative risk of extinction faced by any species. This allows scientists to study changes over time to generate an overall index of change. This information will be used as one of the indicators to measure progress towards Target 12 of the Convention on Biological Diversity Strategic plan for 2020 - 'the extinction of known threatened species has been prevented and their conservation status, particularly of those most in decline, has been improved and sustained'. The Index also has a potential use with other global targets, such as the UN Sustainable Development Goals.

With a global estimate of 391,000 vascular plant species, a comprehensive assessment of every species, repeated over 5 or 10 year timescales, would be impractical. A sampled approach was therefore adopted. All known gymnosperms were included (<1,500), complemented by a random sample of 1,500 species drawn from each of the other groups to be treated for the SRLI for plants: bryophytes, pteridophytes, monocots and legumes – about 7,000 species.



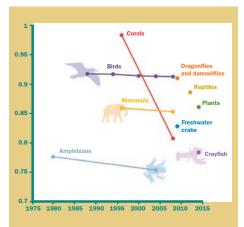
Proportion of species estimated to be in each extinction risk category. CR – Critically Endangered; EN – Endangered; VU – Vulnerable; NT – Near Threatened; LC – Least Concern; DD – Data Deficient (RBG Kew 2012)

During Phase 1 (2006–2010), IUCN Red List assessments for the sampled species were prepared, using herbarium specimen data, GIS analysis, published literature and, where available, expert input. Kew scientists and colleagues at the Natural History Museum, London, worked closely with IUCN specialist groups to complete the assessments.

'The results of the first SRLI for plants showed that 20% are threatened with extinction.'

The results of the first SRLI for plants showed that 20% are threatened with extinction - in the top three IUCN categories of Critically Endangered, Endangered or Vulnerable. A further 8% are classified as Near Threatened, which means that they are not yet threatened but may become so without conservation actions. The greatest threats to plants are driven by human pressures. The conversion of natural habitats to agricultural use, for example, directly impacted 33% of threatened species. Compared to the Red List Index for other groups, plants are more threatened than birds, equally as threatened as mammals, but less threatened than amphibians.

We now have a snapshot of the current status of plant diversity but, to monitor trends, we will need to reassess these species. The new team at Kew will work with its partners to gather new information and reassess the sampled species. We would welcome any information on these species – a full list is available on our website www. threatenedplants.myspecies.info. If you have expert knowledge in a particular flora or country we would also welcome your help as reviewers for these new assessments.



A value closer to 1 indicates most species in the group are at low risk, a value towards 0 indicates species that are moving towards extinction. Values for crayfish, freshwater crabs, dragonflies and damselflies, reptiles and plants are based on a sampled approach.

Reproduced with permission from Brummitt et al. (2015)

Red List Index value of species survival for plants in comparison with other groups (RBG Kew 2016)

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State of the World's Plants website:

https://stateoftheworldsplants.com

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Long-term preservation of recalcitrant seeds

DR DANIEL BALLESTEROS (Tree Seed Science Research Fellow for Europe, Weston Global Tree Seed Project, RBG Kew), DR ANNE VISSCHER (Career Development Fellow - RBG Kew) and ANDREW WEBSTER (MSc student - RBG Kew)

ong-term preservation of seeds at the standard conditions of seed banks (dried to low moisture contents and stored at low temperatures) is only feasible for orthodox (desiccationtolerant) seeds (FAO 2014). However, a large number of species cannot be preserved under these conditions, as is the case for species producing desiccation sensitive (recalcitrant) seeds.

About 20–25% of the seeded plants of the world produce non-orthodox seeds. But. since the proportion of recalcitrant seeds is habitat dependent, recalcitrance acquires importance in particular habitats, such as tropical evergreen rain forests. Here it has been hypothesized that c.47% of the indigenous flora may produce recalcitrant seeds (Tweddle et al. 2003; Daws et al. 2006).

'Cryopreservation... a potential long-term storage solution for these species'

Cryopreservation - the process of using subzero temperatures to preserve living cells and tissues - offers a potential long-term storage solution for these species (Walters et al. 2013; FAO, 2014; Pritchard et al. 2014). The Comparative Seed Biology group of the Department of Comparative Plant and Fungal Biology at Kew has a long history of cryobiology research. Additionally, they are currently testing whether there are any molecular solutions for the ex situ storage of recalcitrant seeds. Two researchers from this research group, based at The Welcome Trust Millennium Building, update us on their work in this area.

Advanced research on cryopreservation of recalcitrant seeds. The Global Tree Seed Project

DANIEL BALLESTEROS (Comparative Seed Biology group)

Although cryopreservation appears to be the most feasible option for the long-term storage of recalcitrant seeds, only a small number of species have well-established and successful cryopreservation protocols. This is partly because desiccation sensitivity and responses to cryopreservation procedures

are variable, and need to be ascertained per species as well as genotype (Walters et al. 2013; FAO 2014).

Under the Weston Global Tree Seed Project - Europe, funded by the Garfield Weston Foundation, the desiccation sensitivity of various recalcitrant seeds and their response to innovative cryopreservation protocols, such as vacuum infiltration vitrification, is being investigated (Nadarajan and Pritchard 2014; Pritchard et al. 2014).

A diversity of oak species (Quercus sp.) of Atlantic and Mediterranean origin, as well as other common species of European forests producing recalcitrant seeds, e.g. sweet chestnut (Castanea sativa) and horsechestnut (Aesculus hippocastanum), will benefit from this research. First results are promising, and we aim to establish successful cryoprocedures for the long-term preservation of these iconic and important species.

Project: Molecular solutions for the ex situ storage of recalcitrant seeds

ANNE M. VISSCHER and ANDREW WEBSTER (Comparative Seed Biology group)

In general, tolerance to desiccation in orthodox seeds is enabled by sugars that replace water and form glasses, proteins that help to stabilize macromolecules and membranes, and antioxidants that prevent excessive damage by reactive oxygen species (Alpert, 2006; Berjak 2006; Gaff and Oliver 2013).

Our project will use vacuum infiltration (Nadarajan and Pritchard 2014) to increase the levels of sucrose, glutathione and synthetic Late Embryogenesis Abundant (LEA) proteins in recalcitrant seed embryos, prior to desiccation treatment, storage and viability assessments. The LEA proteins selected for this study are known to be present in orthodox seeds, but either absent or strongly reduced in desiccation sensitive seed tissues, indicating that these proteins may be critical to seed desiccation tolerance (Delahaie et al. 2013).

Our hypothesis is that proteins produced by selected LEA genes present in orthodox seeds, amplified using bacteria in the lab as expression hosts, will lead to improved desiccation tolerance of recalcitrant avocado



Quercus robur embryonic axes growing in vitro.

Persea americana.



Longitudinal section of Aesculus hippocastanum explants used for cryopreservation showing the root and shoot embrvonic axes.

and lychee embryos when they are applied to these embryos using vacuum infiltration. If this is confirmed, it may open up, for the first time, a distinctly useful approach to storing recalcitrant seed embryos under regular seed bank conditions.

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NEWS Kew's Heritage Trees

JOANNE YEOMANS (Gallery Assistant, RBG Kew)





ast year, the Shirley Sherwood Gallery of Botanical Art hosted *Kew's Heritage Trees* – an exhibition of paintings by Kew artist Masumi Yamanaka, trees began four years later, when she painted the Indian horse chestnut. Aesculus indica 'Sydney Pearce'. She was awarded a Royal Horticultural Society (RHS) Gold Medal of Botanical Art, and was inspired to begin

History of the living collections at Kew

Many trees have been added to Kew's



In 1871, Joseph Hooker planted 1,200 conifers in a new pinetum. The Royal Family has planted many trees over the last 100 years, and a ten-year planting programme began in 2004, when mature trees were added to Pagoda Vista.

A record of Kew's trees

Masumi's paintings act as a record

castaneifolia, is the biggest tree at Kew in terms of volume, and its painting was one of the highlights of the show. Kew's specimen, likely to have been planted by Sir William Hooker in 1846, is the largest of its kind in Britain – in 2007 it measured 34 metres high,

The history of the living collections in Kew's Library, Art and Archives

are photographs including one of the Temple of the Sun, built in 1761 by Sir William Chambers. The photo shows the temple

The first State of the World's Plants Symposium

EMMA WILLIAMS (Species Conservation Assessor, RBG Kew)



90 academic institutions attended the inaugural State of the World's Plants Symposium (SOTWP) at Kew. Lord Gardiner, Minister for the Department of Environment, Food and Rural Affairs, opened the symposium, which was accompanied by the launch of the SOTWP report highlighting the global status of plants. There were six themed sessions: climate change; protected

Samara 15

Adapting agriculture to climate change project: Training

ROBERTA HOPE (Science Administrator - Education [Projects], RBG Kew)

Iready in 2016, 13 partner institutions have been trained to collect, handle and conserve seeds of crop wild relatives as part of the Adapting Agriculture to Climate Change project.

The first training was hosted by the Malaysian Agricultural Research and Development Institute in February, and brought together 21 seed conservationists from across Malaysia, Nepal, Pakistan and Vietnam.

The second course was hosted by the Institute of Genetic Resources in Azerbaijan, and included trainees from the Georgian Institute of Botany and the National Botanic Garden of Georgia.

June's African Seed Conservation Techniques course gathered scientists, all working on species of crop wild relatives, from Nigeria, Sudan, Uganda, Ghana, Ethiopia and Kenya.

All courses used lecture, laboratory and fieldwork sessions to cover different aspects of seed conservation, including population



Learning to take herbarium specimens during the Malaysian training course.

assessment, capturing genetic diversity and techniques for processing seeds.

Between them, these institutions plan to make around 1,300 seed collections from crop wild relatives. This vital training helps to ensure that all institutions can follow the MSBP's Seed Conservation Standards, ensuring that collections are of the highest value to future work on adapting agriculture to climate change.



testing wild banana to examine the contents.

Wakehurst's Wild Wood Weekend

IAIN PARKINSON (Conservation and Woodlands Manager, Wakehurst Place, RBG Kew)

The threat to our woodlands has never been greater. Pests and diseases, development, conversion to forestry, mismanagement and neglect are all taking their toll. Despite their rough treatment, our woodlands remain an enduring and endearing feature of the countryside. They provide spaces for learning and recreation, bring benefits for our emotional wellbeing and contribute to social cohesion. They are unique places with complex communities of plants, animals and fungi.

All these unique qualities were celebrated during Wild Wood Weekend over the May bank holiday at Wakehurst, RBG Kew's country estate. Held in beautiful hazel coppice woodland carpeted with bluebells, the event created a market place for weavers, bodgers (wood-turners), charcoal burners and many others showcasing their skills.



Children playing at Wildwood Weekend.

As well as the traditional crafts on show, the festival also explored more contemporary uses for wood and provided visitors with the chance to try tree climbing, bush craft skills or cooking on an open fire. Theatre in the tree canopy brought the woodland story



heatre in the canopy.

o life, and the opening of a new woodland play area encouraged visitors to immerse hemselves in the wilder side of Wakehurst

www.kew.org/visit-wakehurst/whats-on/wild wood-weekend

New MSB agreements

Country	Counterpart Name	Start Date	Duration (Years)
Armenia	Institute of Botany of the National Academy of Science of the Republic of Armenia	April	1
Australia	Botanic Garden and Parks Authority, Western Australia	March	5
Australia	Royal Botanical Gardens Board, Victoria, South Australia	March	5
Azerbaijan	The Institute of Botany	April	3
Chile	Agricultural Research Institute	Мау	5
South Africa	South African National Biodiversity Institute (SANBI)	May	3
Tanzania	Ministry of Natural Resources and Tourism	January	5
Uganda	National Agricultural Research Organisation (NARO)*	February	5

* denotes new partner for the MSBP

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MSB Dashboard			
Total Collections	78,845		
Number of countries	186		
(+ overseas territories)			
Number of families	337		
Number of genera	5,722		
Number of species	36,895		
Collections despatched	8,127		

Next issue

In the next issue we'll be reporting on the work carried out across the MSBP in relation to alpine flora. We're particularly keen to hear from anyone who would like to share their alpine-related work with the wider MSBP community.

Tales from the field

As fieldwork is an important part of all our work, we'd love to feature more stories and photos from your own trips away from the office – do send your contributions to our editorial team.



We want to hear from you!

Samara is your newsletter, so please contact us with any ideas or articles you feel would be of interest to the MSBP.

Samara Editors

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Samara provides information and inspiration for MSBP partners and a flavour of the success of the Partnership. It is available as a PDF from the MSBP website at kew.org/samara

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