Rescue and conservation of a rediscovered endemic fern: *Anogramma ascensionis* on Ascension Island

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Abstract

The Ascension Island endemic fern, *Anogramma ascensionis*, was listed as extinct on the 2003 IUCN Red List, having been last seen 1958. A 2009 survey rediscovered four plants on a remote cliff on the Island's highest peak, Green Mountain. Spores collected from two of these plants were cultured *in vitro* at the Royal Botanic Gardens, Kew, where a living collection of thousands of gametophytes and hundreds of sporophytes has been developed. Initial results from liquid nitrogen storage indicate that this may be a viable long-term storage option for both spores and gametophytes.

Prior to the large-scale invasion of the Island by alien species in the 19th and 20th centuries, which dramatically transformed Ascension's vegetation, it is likely that *A.ascensionis* flourished in sheltered, humid and shaded parts of the mountain. These habitats have been very heavily invaded, especially by non-native maidenhair ferns, *Adiantum* spp., providing an indication of why *A.ascensionis* came so close to extinction. We have developed a multidisciplinary approach to species recovery, involving *in vitro* culture, up-grading the Green Mountain nursery and providing horticulture training, the clearing of invasive species and controlled translocation, as the road map for re-establishing functional populations of this critically endangered island fern.

Keywords

GSPC; *ex situ* conservation; horticulture; Red List; threatened plant; UK Overseas Territories;

Introduction

Ascension Island lies in the middle of the South Atlantic Ocean, 1,500km from the coast of Africa, just over 2,000km from South America and 7,500km from the UK. Ascension is a UK Overseas Territory, one of 16 former colonies that remain part of the UK. Kew has an active programme providing botanical support to help the territories implement the Global Strategy for Plant Conservation (GSPC) (Clubbe *et al*, 2010).

Ascension is a small (97km^2), young (1Myr old) volcanic land mass with a hot, dry climate, except for the moister upper slopes of Green Mountain which rises to an elevation peak of 859m. Green Mountain is Ascension's only National Park, established in 2005 (Ascension Island Government, 2014a) and the habitat for its most important plant diversity (Figure 1). Being a young volcanic island with poorly formed soils and a relatively short time for plant colonisation, Ascension supports a relatively modest vascular flora. A comprehensive island-wide census in 2009 recorded 249 wild species, 95% of which are considered non-native, but of the native flora more than half are endemic to Ascension (Lambdon & Darlow, unpublished data). The biggest current threat to Ascension's native flora is non-native invasive species. Results from the 2009 census suggested that non-native species occupy approx 99% of the vegetated area of Ascension.

One of the most important plants on Green Mountain is the endemic Ascension Island parsley fern, *Anogramma ascensionis* (Pteridaceae) which we know from the type specimen in the Kew herbarium; it was collected by Kew's former Director Sir Joseph Hooker in 1843 (UKOTs Online Herbarium, 2014) (Figure 2). The only confirmed 20th century record for *A. ascensionis* is by Eric Duffey in 1958, although with no associated voucher specimen (Duffey, 1964). The Ascension Government Conservation Department conducts an annual census lead by Ascension's Conservation Officer, Stedson Stroud, and despite concerted efforts no *A. ascensionis* had been

found at the site documented by Duffey nor in any other likely habitat locations. As a result of these directed, unsuccessful searches over several years, *A. ascensionis* was assessed as Extinct for the 2003 IUCN Red List (Gray, 2003). The annual plant censuses continued and in 2009 four small plants of *A. ascensionis* were found on a remote cliff side on Green Mountain, and a race against time was started to save this fragile species. Plants were monitored and watered in the field to ensure survival and to induce sporulation (Figure 3). Sterile collecting materials were sent to Ascension from Kew and when the spores were ripe the fronds were collected into sterile petri dishes and with the support of the Island's Administrator and the Royal Air Force we were able to get the spores from the field into micro-propagation in the laboratory at Kew within 24 hours (Gill, 2010).

At Kew the team in the Conservation Biotechnology Unit successfully established these spores in micro-propagation and have taken the fern experimentally through its whole life-cycle, through to mature sporophytes weaned out onto growing media in pots in the Tropical Nursery and collected viable spores from the mature sporophytes. Under these optimal conditions the sporophytes are substantially larger that any seen in the wild on Ascension but interestingly of a size similar to Hooker's original collections, giving the impression that the re-discovered plants are growing in sub-optimal conditions (Figure 4). We now have hundreds of viable plants in *ex situ* conservation at Kew. The Ascension conservation team have been to Kew for training in micro-propagation techniques as well as in the conventional propagation of ferns. Kew horticulturists have been to Ascension to help upgrade the Green Mountain nursery. Capacity building is a very important aspect of Kew's approach to partnerships and a key contribution to target 15 of the GSPC (CBD, 2011)

Recently gametophytes and spores have been stored and successfully recovered from liquid nitrogen and grown on agar right through to the sporophyte stage, providing a viable long-term *ex situ* storage solution for this threatened fern. The whole protocol has been documented, shared with the Ascension team and published as a contribution to GSPC Target 3 (Baker *et al*, 2014).

Targeted surveys in 2009-2012 discovered three further populations around Green Mountain, giving us encouragement that *Anogramma* is more widespread but cryptic and hanging on in the more inaccessible areas of Green Mountain. More worrying is that two species of highly invasive maidenhair ferns (*Adiantum raddianum* and *A. capillus-veneris*) are rapidly spreading on Green Mountain and occupying *Anogramma's* favoured niches, the wetter south and west as noted by Hooker. As a result of this re-discovery and its securing in *ex situ* conservation, *A. ascensionis* was re-evaluated and down-listed from Extinct to Critically Endangered for the 2010 IUCN Red List, but its existence in the wild is still fragile (Lambdon *et al*, 2010). Work is well advanced to upgrade the facilities at the Green Mountain nursery to provide better growing conditions for Ascension's endemic plants including *A. ascensionis* (Figure 5). The first batches of flask-grown plants have been repatriating from Kew to Ascension and are growing well in the Green Mountain nursery. The challenge now is to develop a successful protocol for weaning the plants from flasks into a local growing medium and maintaining them in the Nursery until they are at a suitable stage for re-introduction. This process has started and the advantage of having lots of material now available is that a range of approaches can be tried in an experimental way in Ascension in order to optimise the protocol.

The challenge for re-introduction is to tackle the invasive species that dominate *Anogramma's* habitat and in particular the two species of non-native, invasive maidenhair fern (*Adiantum raddianum* and *A. capillus-veneris*) which aggressively out-compete *A. ascensionis* for its favoured niches (Baker *et al*, 2014) (Figure 6). Some experimental hand weeding of potential re-introduction sites has been undertaken as part of the preparations for re-introduction.

An added complication to re-introduction has emerged whilst studying the life cycle of *A. ascensionis* at Kew. Unusually for ferns, the sporophyte seems to be an annual with green spores which have limited viability. The evidence points towards the sporophyte being the short-lived part of the life cycle with the gametophyte being the persisting generation, living for several years

cryptically in the volcanic substrate (Baker *et al*, 2014). In the light of this, more success may be gained by re-introducing the gametophyte rather than the sporophyte, an approach which we are currently investigating.

A Species Action Plan was completed for *A. ascensionis* in 2009 as part of a more comprehensive conservation plan developed for Ascension's native flora (Lambdon *et al*, 2009). In light of these new discoveries and a better understanding of the biology of *A. ascensionis*, this Species Action Plan is being revising as part of the development of a more comprehensive Biodiversity Action Plan for Ascension currently being funded by the UK Government's Darwin Initiative Programme (Ascension Island Government, 2014b).

The Ascension Islanders have really taken *A. ascensionis* to their hearts and it has attracted lots of publicity and community engagement locally. There are regular articles in Ascension's weekly newspaper, *The Islander*, and a special issue of stamps to celebrate the re-discovery of this thought extinct endemic fern was issued in 2011. Constitutionally, Ascension is a dependency of St Helena and as such used the flag of St Helena. However, in 2013 Ascension got its own flag for the first time which was first raised on Ascension Day, 11 May 2013. Significantly *A. ascensionis* is represented on this flag. Is this the first time an endemic fern has made it onto a national flag? An interpretation board recently erected on Elliot's Path, a circular walking path in Green Mountain National Park, tells the story of the re-discovery of *A. ascensionis* and highlights the conservation work underway on Ascension; this is a contribution to Target 14 of the GSPC (CBD, 2011).

In summary this conservation success story highlights how botanic gardens – using their unique skills set comprising science, horticulture, education and awareness raising – can be brought together in a multi-disciplinary way to save species and deliver successful conservation.

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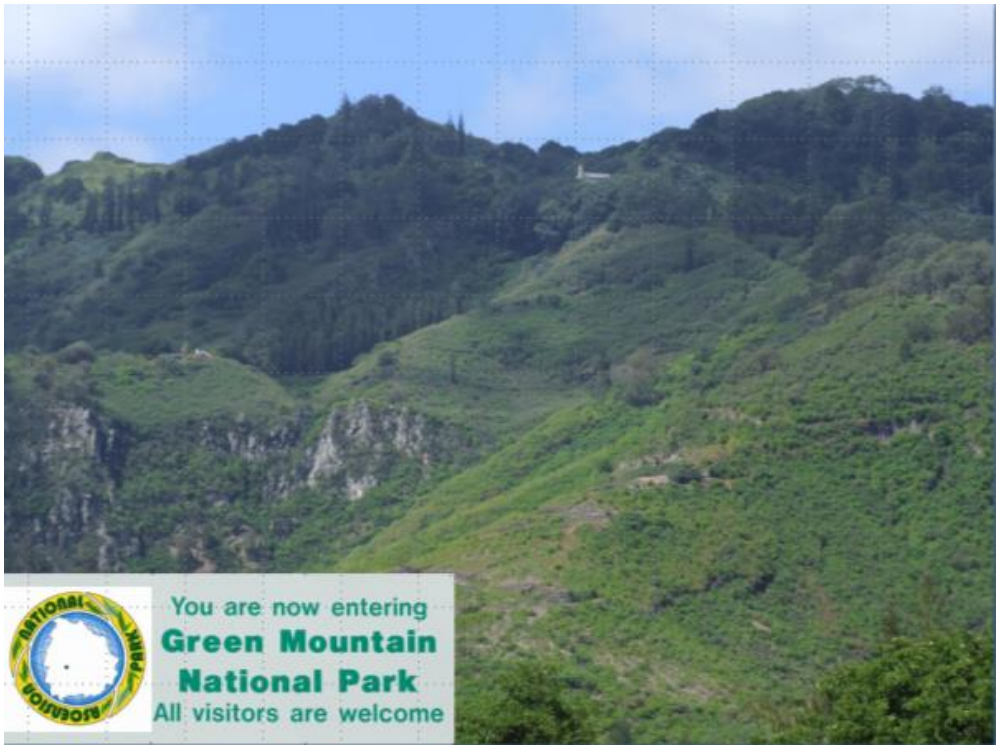

Figure 1: Green Mountain National Park (photo: Colin Clubbe)

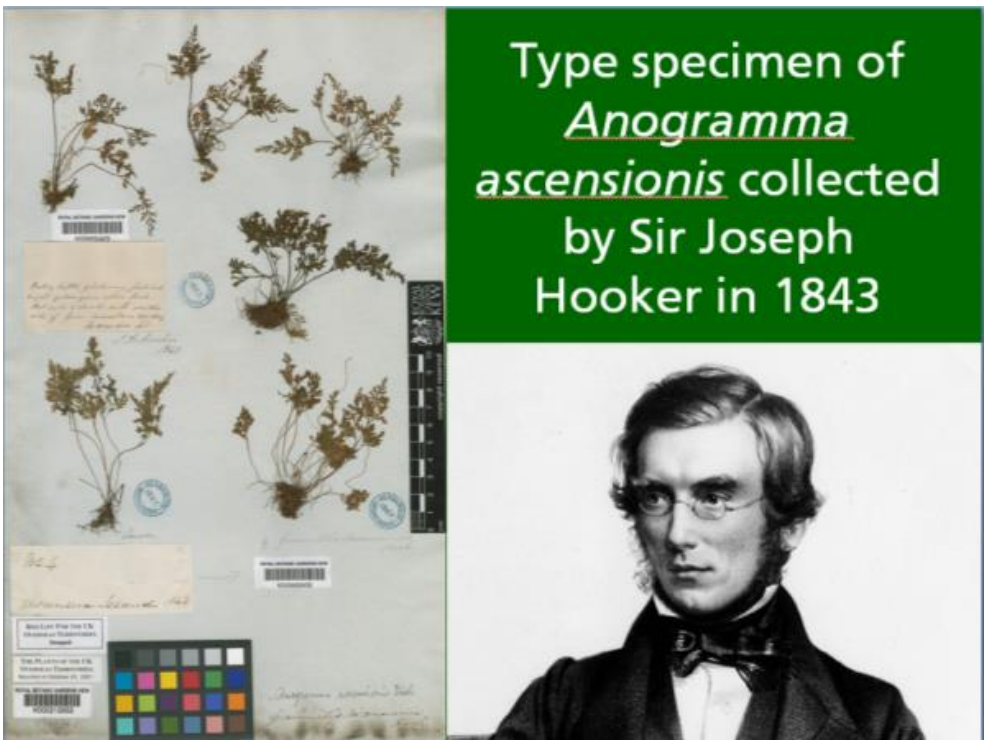

Figure 2: Type specimen of *Anogramma ascensionis* collected by Joseph Hooker in 1843 and an image of Joseph Hooker from the Kew archives. Image copyright: Trustees of the Royal Botanic Gardens Kew.

Figure 3: *Anogramma ascensionis* on Green Mountain.

Clockwise from left: Young *Anogramma ascensionis* sprorophyte (photo: Colin Clubbe); Stedson Stroud examining *A. ascensionis* (photo: Colin Clubbe); Olivia Renshaw watering *A. ascensionis* (photo: Phil Lambdon).

Figure 4: *Ex situ* cultivation of *Anogramma ascensionis* at Kew.

Clockwise from left: Cultures of *A. ascensionis* gametophytes in the Conservation Biotechnology Unit at Kew (photo: Andrew McRobb); Mature sporophyte of *A. ascensionis* in the Tropical Nursery at Kew (photo: Andrew McRobb); Hooker's type specimen with cultures of *A. ascensionis* at different stages (photo: Colin Clubbe); Transferring *A. ascensionis* gametophyte cultures from plates to flasks (photo: Andrew McRobb).

Figure 5: The Green Mountain Nursery in Ascension (photo Colin Clubbe).

Figure 6: Invasive *Adiantum* spp. rapidly colonise the volcanic substrate on Green Mountain, out-competing *Anogramma ascensionis* (photo Colin Clubbe)