Darwin Initiative – Final Report

*(To be completed with reference to the Reporting Guidance Notes for Project Leaders (http://darwin.defra.gov.uk/resources/reporting/) - it is expected that this report will be a **maximum** of 20 pages in length, excluding annexes)*

Darwin project information

Project Ref Number	EIDPO13
Project Title	Integrating Evolutionary History and Phylogenetic Measures of Biodiversity into Conservation Planning
Country(ies)	South Africa
UK Contract Holder Institution	Royal Botanic Gardens, Kew (RBG Kew)
UK Partner Institution(s)	
Host country Partner Institution(s)	South African National Biodiversity Institute (SANBI) University of Cape Town (UCT) University of Johannesburg (UJ)
Darwin Grant Value	£98,297
Start/End dates of Project	01 August 2006 to 31 July 2008
Project Leader Name	Dr Vincent Savolainen (now dual appointee between The Royal Botanic Gardens Kew and Imperial College London)
Project website	http://www.sanbi.org/research/dnabank.htm (original project website)
Author(s), date	Dr M. Powell & Dr V. Savolainen, 30 November 2008

1 Project Background

This post-project (hereafter referred to as the 'project') results from a previous successful Darwin project, 162/12/008: DNA banking, phylogeny and conservation of the South African flora. The original project established the first DNA banking facility in South Africa, which archived genetic material from at least one species of 1,237 South African angiosperm genera (the total number of DNA extracts now stored in the DNA bank stands at 5,176), and the required legal agreements were put into place to allow material transfer to and from the DNA bank, ensuring appropriate benefit sharing of these genetic resources. Based on genetic material from the DNA bank a phylogenetic 'tree of life' was produced for the Cape flora, comprising 735 of the 943 genera currently recognised within the Cape, and was used to identify areas of endemicity and high priority for conservation (results reported on previously, Forest *et al*., *Nature*).

As detailed in the previous annual reports, this project seeks to build upon the successful data production and networking of the original project by focusing on four scientific aspects deserving further attention:

- link conservation planning with the phylogenetic data (some of which were produced during the original project), by coordinating follow-up scientific research;
- calculate extinction risks, building on red lists for the South African flora;

- continue to transfer knowledge regarding the use of phylogenetic data to in-country scientists, students and conservationists, by providing training and research opportunities;
- provide baseline data for the development of future conservation actions within current partnerships and to develop new partnerships (e.g. see below and with other Darwin projects in South Africa), and extend the use of DNA resources to DNA barcoding for conservation (e.g. at the Kruger National Park; KNP).

Outstanding project achievements include:

- The identification of a universal DNA barcode for flowering plants (published in the *Proc. Natl. Acad. Sci. USA*; with the subsequent application of patenting this discovery in the US, entitled 'DNA Barcoding of Plants' P41629US);
- The successful award of a five-year Royal Society/South African National Research Foundation capacity building grant to continue working on biodiversity, conservation and DNA barcoding of South African flora;
- The appointment of host country partner Dr van der Bank as Tree-BOL's 'Regional Working Group Co-Chairperson for Africa', giving her responsibility for coordinating and reporting upon all of the DNA barcoding activities which are being undertaken in Africa;
- A proposal has been submitted to the South African NRF for the molecular systematics laboratory at UJ, headed by Dr van der Bank, to be recognised as a Centre of Excellence (CoE) in DNA barcoding.

2 Project support to the Convention on Biological Diversity (CBD)

This project assists South Africa in meeting its obligations under the Convention of Biodiversity; its relevance is spread across many articles and cross-cutting issues, and the breakdown of these is provided in Annex 3.

The project contributes mostly to nine of the articles under the CBD (see Annex 3) and especially articles 6: General Measures for Conservation and Sustainable Use; article 7: Identification and Monitoring; article 12: Research and Training; and article 15: Access to Genetic Resources. The establishment of DNA banking facilities at SANBI Kirstenbosch and UJ has facilitated research on biodiversity and conservation (article 6), through the generation of molecular phylogenetic data, which have subsequently enabled conservation assessments to be made using measures such as phylogenetic diversity (PD). The PD analyses and conservation assessments of the Gouritz region in the Little Karoo, Kruger National Park (KNP) and the Cape floristic region have strong potential for aiding decision-making processes with conservation activities in South Africa, by supplying an additional tool for identifying areas of future conservation efforts.

Maintenance and organisation of data relevant to identifying and monitoring components of biological diversity (article 7) has been enhanced by the storage of genetic material in the DNA banking facilities at SANBI and UJ, both these databases are available online (original project website and www.florakrugerpark.org/. Identification of South African flora has been made facilitated by the research on DNA barcoding and the phylogenetic data produced for the Cape flora, coupled with the increase in collections at national herbaria. The use of PD and extinction risk analyses has aided the identification of regions requiring urgent conservation action. The CBD vision for 2006 (e.g. CBD press release at http://www.biodiv.org/doc/press/2006/pr-2006-01-cbd-en.pdf) highlights priorities in addressing the 2010 biodiversity target; both the use of DNA barcoding and PD for rapid assessment of biodiversity hotspots are proving to be highly valuable with respect to monitoring the CBD's 2010 biodiversity targets.

Research and training activities (article 12) have been at the forefront of both the original project and the post-project. The original project trained 39 students in molecular biology techniques over the course of four week-long training courses, whereas the post-project has trained 66 research staff and students (both post-graduate and undergraduate) in a range of research skills (see Section 4.3.2 below).

The project has contributed towards access to, and the protection of, genetic resources within South Africa (article 15); in particular the modification of SANBI's material transfer agreement to allow DNA samples to be passed to outside institutions through the DNA bank has been a significant step forward. In terms of the Biodiversity Strategy and Action Plan it ensures that spatial conservation assessments can use phylogenetic information in future assessments. The establishment of the DNA banks at SANBI and UJ is recognised as an important factor in helping South Africa meet its obligations under the CBD, as Action Plan 50.4 for South Africa targets the number of genomes stored in DNA banks as an indicator of the sustainable use of biological resources and the equitable sharing of the benefits (CBD target 3.1).

In addition, SANBI works closely with the CBD National Focal Point in RSA and there is very good communication with government regarding SANBI's role and activities.

3 Project Partnerships

Two Memoranda of Understanding were agreed upon and signed between both RBG Kew and UJ, and RBG Kew and UCT, within which strict rules and guidelines pertaining to the transfer of material and running of the project were outlined.

The working relationship between UK and South African project partners has continued to be very fruitful and productive, with regular communication between institutions and visits from project members to both the UK and South Africa, e.g. (excluding specific training activities, see Section 4.3.2):

- A pre-project meeting was organised in July 2006 in South Africa at the South African Society of Systematic Biology (SASSB) conference, which was attended by several project partners;
- Drs Savolainen, van der Bank, and Powell went to the KNP for a workshop on DNA barcoding and collected new samples for the project in January 2007;
- Drs Savolainen, Verboom and Dreyer met in January 2007 to organise the publication of a special theme issue of *Molecular Phylogenetics and Evolution* on the phylogenetics of the Cape biota (due to be published in early 2009);
- Dr van der Bank visited project partners at RBG Kew in July 2007 and January 2008;
- Drs Savolainen and Powell (the latter appointed UK Darwin Project Officer in October 2007) visited Prof Hedderson and Dr Verboom (UCT), Dr Pauw and Dr Dreyer (University of Stellenbosch) and Dr van der Bank (UJ) in August 2007;
- Mr van Alphen Stahl (appointed as extinction risk analyst at UCT) worked with UK project partners at Imperial College London on the PRECIS database in December 2007;
- Dr Powell visited the two project-funded MSc students at UCT in March 2008 to discuss the progress of their projects.

Collaboration between partners of a further Dr Savolainen-led Darwin project (14-001: Conservation and Monitoring of Meso-American Orchids) has been highly productive and resulted in a key publication on DNA barcoding (see Section 4.5).

The successful working relationship between South African and UK project partners is further reflected by the award of a five-year Royal Society/South African National Research Foundation grant to work on 'Regional patterns of biodiversity and conservation in South Africa: the flora of the Kruger National Park as a case study' (£228,451 + ZAR 1,593,642 over 5 years).

Links between project partners, particularly Dr Michelle van der Bank (UJ), and SANBI, UCT, SAAB (South African Association of Botanists) and SASSB (South African Society for Systematic Biology) have been enhanced during the project's lifetime, with the UJ molecular laboratory being utilised not only by different research groups within UJ, but also by collaborators at SANBI (Pretoria and Kirstenbosch) and UCT.

Collaboration with Prof Gideon Smith and Yolande Steekamp (SANBI Pretoria) has also been strengthened as a result of data sharing with the PRECIS (Pretoria National Herbarium) database, and an Extinction Risk Analysis course held in July 2008.

Gouritz conservation assessments have been initiated by Dr Felix Forest (Kew) and Prof Richard Cowling (Nelson Mandela Metropolitan University).

The SAAB/SASSB VII conference in January 2008 produced a special themed issue of *Molecular Phylogenetics and Evolution* which cemented the partnership between this project and those two South African societies.

Partnerships have also been established and strengthened with major groups heading DNA barcoding consortia (i.e. the CBOL and Tree-BOL; see Section 4.3.5 for further details).

4 Project Achievements

4.1 Impact: achievement of positive impact on biodiversity, sustainable use or equitable sharing of biodiversity benefits

The project has provided staff training and technology transfer, which will help to ensure that the capacity remains in the host country to take an active role in the conservation and sustainable use of biodiversity: a total of 66 staff and students have received specific training in areas such as molecular biology techniques, extinction risk analyses, and phylogenetic diversity and conservation over the duration of the post-project. In addition to this, two MSc and three Honours studentships (supported by the project) have been completed.

Plant genetic resources are made available for scientific research: The sample collections undertaken as part of the project have enhanced the DNA-bank databases of the molecular systematics laboratory at UJ, which now comprises approximately 2,500 samples, and SANBI Kirstenbosch (>5,000 samples). All databases are available online. The collecting effort has also increased the UJ herbarium collections, which now houses more than 1,000 herbarium sheets, duplicated at the herbarium at Skukuza (Headquarters of the KNP).

Under the National Environmental Management Biodiversity Act (NEMBA, Act No. 10 of 2004) SANBI has been mandated by the South African government to co-ordinate and promote research into taxonomy, and indigenous biodiversity, and to monitor and report on the status of the country's biodiversity. In addition, SANBI is responsible for monitoring and reporting to the Minister of Environment on the conservation status of all listed threatened or protected species and ecosystems, with the aim of aiding the Minister in biodiversity planning. This project has assisted the fulfilment of this mandate by making use of, and expanding upon, the already existing DNA database, DNA bank, and continuing phylogenetic studies to better understand the taxonomy of South African flora. Furthermore, this project has provided, through the use of phylogenetic diversity (PD) analyses and training in extinction risk analysis, a direct assessment of the conservation status of species and ecosystems, thus providing a source of information which SANBI can draw upon to meet the government mandate outlined in NEMBA. The original project was clearly endorsed in the South African 2nd National CBD Report, especially with regard to Article 7 and Article 9, e.g. *"Together with the Kew Royal Botanic Gardens, the NBI [SANBI] has established a DNA Bank in South Africa. It will represent a unique archive of plant genetic diversity in South Africa, holding over 2 200 genomes from all genera. It will also serve as a resource to facilitate the discovery of novel genes and for the identification of areas of high priority for conservation.", and we believe that this post-project has continued in this way.*

The high levels of within-country and international publicity which resulted from a PD assessment of the Cape flora (reported in Forest et al., Nature), coupled with SANBI's association with the South African Minister of the Environment and the subsequent conservation assessments of the KNP and Gouritz, increase the likelihood that the findings of this research will have an impact on conversation decisions within South Africa. The Forest *et al*. paper resulted in a joint press release between RBG Kew, UK Defra/Darwin Initiative and SANBI publicising our work, and was cross-referenced with a News & Views by Nature about the paper. Several articles have reported on our results, including several interviews with project partners (See section 4.3.3 for details). There are always sceptics about using PD for practical conservation, but South Africa is one of the few countries that has really taken science

into conservation planning, thanks to eminent conservationists such as Professor Richard Cowling. Our research will contribute to this national effort.

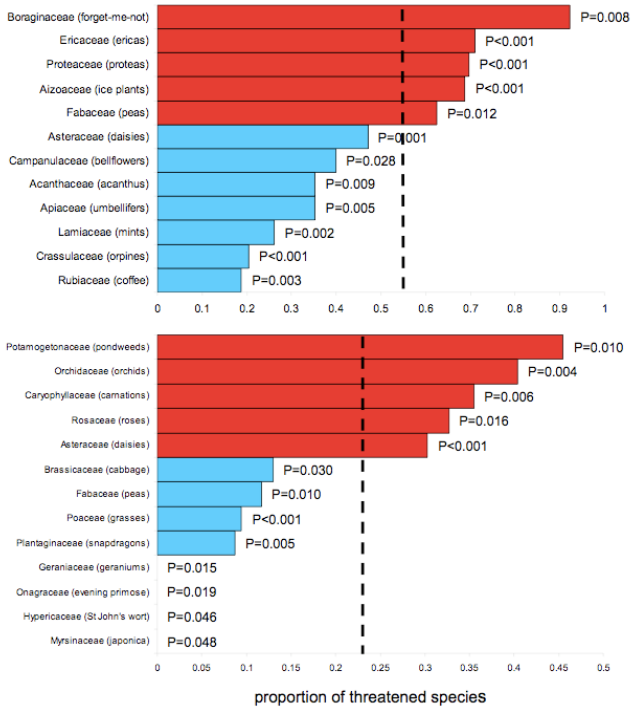
4.2 Outcomes: achievement of the project purpose and outcomes

The post-project purpose, extending upon the goal of the original project to establish a DNA bank of archived plant genetic resources within South Africa, was to build upon the data compilation and scientific networks established in order to address four scientific aspects which were deserving of further attention: i) co-ordinate research; (ii) calculate extinction risks; (iii) transfer knowledge to in-country scientists, students and conservationists with regard to the use of phylogenetic data and DNA barcodes; (iv) publish concerted conservation actions. The post-project has ultimately been successful in the achievement of these objectives, as is detailed below.

Considerable research has continued on the data produced on the original project, and co-ordination of this has been carried out by the three principal partner institutions. The results of this research are listed as publications (Annex 4) and have been disseminated to a wide scientific and non-scientific audience (Section 5).

The geographic distribution data derived from the PRECIS database with the assistance and cooperation of Prof Gideon Smith and Yolande Steekamp of SANBI Pretoria, has been upgraded to enable future assessment of extinction risks within South African taxa. The database, an inventory which contains taxonomic and distribution (in Quarter Degree Squares; QDSs) information on all of the angiosperm genera in South Africa (comprising approximately 10, 000 species) had to be updated to match the taxonomic delimitation described by the Angiosperm Phylogeny Group and that of *rbcL* sequence data from GenBank. The database had to be converted from its original format (Access) to a more readily usable style (R computing environment) to enable the assessment of extinction risks to be made.

We conducted a study on plant threat in South Africa, which we also compared to the situation in the UK, using available resources form the post-project and some other unpublished data.



Going beyond the APG family-level classification used in previous studies of plant extinction risk, we compiled a genus-level taxonomy that reflects current phylogenetic knowledge: this dataset encompasses 13,995 genera classified in seven hierarchical clade-levels. We then focused on the UK and the Cape of South Africa, and calculated the proportion of threatened species across these hierarchical levels. Using randomisation tests, we identified several families with an unusually high proportion of species at risk of extinction (10 families), whereas another 15 families had a significantly lower than expected proportion of threatened species (Fig. 1; collaborator T. J. Davies & coordinator Savolainen, unpublished data). These results demonstrate the importance of detailed phylogenetic information.

Figure 1. Preliminary results on extinction risks in plants. Families with higher than expected proportions of threatened species are shown in red, whereas families with significantly lower proportions of threatened species are shown in blue; both in South Africa (top) and the UK (bottom; see text).

We also used species-level trees of the *Moraea* and *Protea*. For each species assessed in the South African Red List, we also compiled soil preference, fire strategy and pollinator data available in the literature. We found levels of threat to be relatively independent of the evolution of traits along the phylogenetic trees, with a few exceptions such as beetle-pollinated species.

In this sense, our pilot study mirrors ambiguous results of the effects of traits on extinction found elsewhere, e.g. in the flora of Singapore, and calls for more comprehensive studies.

In the Cape we also detected significant geographic patterns in the intensity of threat, and a link between rates of diversification and risk of extinction, indicating that PD may play a key role in extinction.

Following on these results, several training activities were organised throughout the duration of the project to provide South African scientists and students with the knowledge and skills required to generate and use phylogenetic data and DNA barcodes. A full list of training activities is provided in Section 4.3.2.

Results of PD and extinction analyses are on their way to being published and form the core of our conservation assessments (see Section 4.3.4).

4.3 Outputs (and activities)

There are five types of project output in our logframe: 1) DNA barcoding; 2) Training; 3) Dissemination; 4) Conservation assessments; 5) South African Conservation Scientists network.

4.3.1 DNA barcoding

The DNA barcoding of the flora of the KNP aimed to identify a universal DNA barcode for angiosperms. This post-project provided a platform for the exchange of information, especially with regard to which DNA region will eventually be used as a barcode. Following protocols established by the Plant Working Group of the Consortium for the Barcode of Life (CBOL) (www.kew.org/barcoding), several genomic regions have been tested on the flora of the KNP. Analyses were carried out on eight potential DNA barcoding regions for over 700 taxa from the KNP (including 55% of the 400 species of trees and shrubs occurring in the KNP), far exceeding the target of producing 250 barcodes, and the *matK* region of the plastid genome was found to be the most suitable. The full DNA barcoding database can be accessed online (www.florakrugerpark.org), and the results of this studied were published to considerable international acclaim (see 4.3.3) by Lahaye *et al*. in *PNAS* in February 2008 (pdf supplied, annex 10).

4.3.2 Training

Training has been a key component of the project, and there have been a range of training activities organised over its duration. In total 66 students and staff have been trained in areas related to the project's research, either in the form of research visits (three such visits took place, totalling 10 person weeks) to the UK by South African project partners, or by structured training courses run in South Africa by the UK partners (four such courses were run totalling 51 person weeks).

Apart from the visits listed in Section 3, a further three research visits to RBG Kew were made by South African project partners specifically to receive training: i) Ms Gopal (DNA bank manager at SANBI) visited the DNA bank managers at RBG Kew to receive further training (2 July to 15 July 2007); ii) Dr Lahaye (UJ) visited RBG Kew (22 July to 18 August 2007) to continue the collaboration between the two institutions by analysing two datasets and writing a paper to identify a universal DNA barcode for the flora of the KNP; and iii) Mr van Alphen Stahl visited project partners at Imperial College London (29 November to 19 December 2007) to work on the valorisation of the PRECIS database for extinction risk analyses.

Training courses

1) Phylogenetic diversity in conservation:
Under the hospice of this Darwin project, Prof Terry Hedderson hosted a course at UCT on the use of phylogenetic diversity in conservation, a topic that is at the heart of our research activities. The week-long course was taught in January 2007 by Dr Richard Grenyer (previously a phyloinformatician at RBG Kew, now based at Imperial College), Dr Felix Forest (RBG Kew, formerly postdoctoral fellow at SANBI/UCT during the original Darwin project), and Dr Dan

Faith (instigator of the use of phylogenetic information and PD in conservation; senior researcher at the Australian Museum in Sydney). The course was attended by an audience of South African students and scientists (18 people from UCT, U Stellenbosch and SANBI) and subsequent evaluations of the course were very positive (once averaged, all course activities were marked above 4 out of a maximum satisfaction of 5). In these evaluation forms we also asked whether the participants would now consider using PD in conservation assessments, and all answered yes. A copy of the training course booklet was given to each student as a technical guide.

2) Laboratory practical in DNA barcoding:
A week-long course was held at UJ on 'DNA barcoding: a practical guide' by Dr Martyn Powell from 7-11 April 2008. Attendees were 16 undergraduate students (five were previously disadvantaged South Africans; Annex 6) and one professor from the university; the course involved introductory lectures to the molecular biology techniques being used, and hands-on laboratory practicals for each student. A copy of the training course booklet and lecture material was given to each student as a technical guide (Annex 7 and 7a).

3) Grant writing:
A four-day course on 'Grant writing' was given to seven participants (Postgraduate students, postdocs and staff from UJ, SANBI Pretoria, and the University of Cape Town; Annex 8) by Dr Vincent Savolainen at UJ from 14-17 April 2008. Training was focused on grant writing, using the Logical Framework approach for project design and management (using previous Darwin Initiative grants as case studies). The course used short lectures, hands-on exercises, and role plays (i.e. asking the attendees to write 'ghost' grants, which will be passed onto another attendee 'for review' and discussed at round-tables).

4) Extinction Risk Analysis:
A two-day course on 'Extinction Risk Analysis' was given at SANBI Pretoria by Dr Richard Grenyer (Imperial College) to 23 participants on 28-29 July 2008 (Annex 9). The theory and practical application of this approach was explained, with break-out groups completing computer-based exercises.

MSc and Honours students

Two Masters students (Alastair Potts and Matthew Britton), whose bursaries were funded by the project, are working at UCT with Prof Terry Hedderson, and they are due to complete their projects by the end of 2008/early 2009. Both are intending to uptake PhD positions in the New Year. The project funded the bursaries of three honours students at UJ working with Dr van der Bank; two of them (Phip Moolman and Genevieve Thompson) completed their six-month molecular biology course and individual projects on "DNA barcoding of the trees and shrubs of the KNP" in December 2007, and a third, Anneli van Rooyen, begun her DNA barcoding project in January 2008 (she should finished at the end of 2008/early 2009). Some of these projects were not directly linked on extinction risks but all built on the DNA resources from the original project and the phylogenetic expertise of our network.

EXAMPLE OF A MSC PROJECT FUNDED THROUGH THIS POST-PROJECT:

The Phylogeography of three plant species in the Little Karoo, South Africa; Mr Alastair Potts, UCT (Expected date of completion: end of 2008)

Abstract: An understanding of the current and historic processes that have affected present-day species distributions is essential for the conservation and management of broad geographic regions. To investigate the phylogeographic patterns within the Little Karoo, three species were studied. Specifically, the three species are *Berkheya cuneata*, *Nymania capensis* and *Pappea capensis*. *B. cuneata* is a Little Karoo and Succulent Karoo endemic, while *N. capenis* is found throughout the Little Karoo and neighbouring Albany Thicket, as well as in a disjunct population in the Eastern Gariep region. *P. capensis* is a widespread species that occurs from the equator through to the Little Karoo. The results show that an inselberg in the middle of the Karoo, the Rooiberg, is an effective barrier to seed dispersal for both *B. cuneata* and *N. capensis*. Both the

Succulent Karoo and Subtropical Thicket areas on either side of this barrier should be considered separate management units. *P. capensis* is shown to have low cpDNA genetic diversity in the Little Karoo suggesting a recent expansion from the Eastern Cape. Populations for both *P. capensis* and *N. capensis* display deep genetic splits between the Little Karoo and the centre of the Albany Thicket in the Eastern Cape. These phylogeographic results form the basis for further comparative phylogeographic studies in the region.

4.3.3 Dissemination

Oral dissemination of the project's research has taken place at several national and international conferences and workshops (see also Section 5), for example Dr Savolainen presented the results of the Cape and Gouritz PD analyses at a symposium dedicated to PD at the Evolution meeting in June 2007 (New Zealand); these results were also presented on behalf of the project by Dr Davies in July 2007 at the Annual Meeting of the Society of Conservation Biology in South Africa; several project partners presented results at the SAAB/SASSB VII conference in South Africa in January 2008. Dr van der Bank presented the results and future plans of the KNP barcoding at various occasions: the Dendrological Society of South Africa (26 July 2008); Tree-BOL 2008, The New York Botanical Garden, USA (01-02 May 2008); the 6th Scientific Network, Kruger National Park, South Africa (April 2008); and the Second International Barcode Conference (of the Consortium for the Barcode of Life), Taipei (September 2007).

There have been several press releases in the popular press to publicise the general purpose of the project and its activities, e.g. in Kew Scientist and UJ magazine (autumn 2007 issue).

In addition, there have been a multitude of press releases (over 50 websites reported on the DNA barcoding research) pertaining to specific project's activities, principally DNA barcoding, and these include coverage in national newspapers in the host country (e.g. The Star, 14 February 2008; Lowvelder, March 2008; Kruger Park Times February 2008; Beeld, 23 April 2008) and magazines (e.g. Landscapes SA, January 2008; DNA Barcoding Herald, 2008; African Geographic, May 2008). A selection of these can be found online (http://www.uj.ac.za/PlantMolecularSystematicsLaboratory/Researchprojects/TreeBOLAfrica/Pressrelease/tabid/13946/Default.aspx). In addition to written press, Dr van der Bank has also held interviews on national radio (RSG, morning talks, 2008; Radio Pretoria, 2008; RSG – Eco-Forum, 2008) and television (SABC News, February 2008).

An example of the trend of opinion voiced about the Lahaye *et al*. publication is detailed below with a quote from the Defra website by Joan Ruddock, Minister for Climate Change and Biodiversity:

> *"This is a great breakthrough that could save many endangered plants. The Defra-funded Darwin Initiative has a reputation for producing real and lasting results and I congratulate everyone involved in this project which could have huge benefits for plant identification and conservation in the future."*

A final project workshop was held at UJ in July 2008, and was attended by project partners from both UJ and RBG Kew/Imperial College. Contact was established with three further, South African-based, Darwin projects with a view to hosting a workshop between the projects to discuss putative collaborations and other matters arising from working in South Africa (14/012: Limbovane Outreach Project: Exploring South African Biodiversity and Change; 15/012: Protecting Key South African Biodiversity Sites through Community-based Conservation; 16/003: Tools, training and research for managing eco-hydrology of Cape flora), but ultimately it was not possible to hold this workshop due to the varying time and fieldwork commitments of the projects, and discussions have occurred by email.

Publications are reported in Annex 4.

4.3.4 Conservation assessments

The conservation assessments produced by the project have focused on:

- Providing conservation planners with information on how to prioritise areas for conservation based on PD, in addition to more traditional measures such as taxon richness and diversity
- Measuring extinction risks in the Cape

These conservation assessments are already published or due to be published as 'traditional' scientific papers.

The assessments have utilised the data produced during both the original project and this project to evaluate the PD of three regions within the hotspots: the Cape itself, the Gouritz corridor and the KNP. These results have then been compared to those obtained using more traditional approaches (e.g. taxon richness) to assess whether preserving species richness might not necessarily protect the processes that are responsible for the presence of rich ecosystems.

The conservation for the Cape flora was published in *Nature* (Forest *et al*, mostly as a result of our original project although analyses have taken place also during the post-project), and received a significant amount of positive publicity (abstract provided in Section 5, pdf annex 11). Another paper on the Cape is in preparation by Davies, Savolainen and collaborators (see 4.2 and Annex 15).

In the Gouritz region of the little Karoo, analyses have focussed on detailed habitats and their floristic composition. Our results show that the most recent vegetation types (i.e. fynbos) have low levels of PD given their generic composition. A correlation was also observed between ecosystem status (with respect to IUCN criteria) and PD levels, with the more threatened areas having lower than average PD values (Forest *et al*., *in prep*., annex 16). This same observation has been made in the Cape (Davies *et al*., *in prep*). These conclusions are in agreement with our paper on the entire Cape (Forest *et al*, Nature) and once again show a decoupling between taxon richness and PD. Taken altogether, these results indicate that efficient conservation needs to take into account different biodiversity metrics if one wants to conserve processes, patterns and richness.

The target of producing a phylogenetic tree of all genera of KNP, and assessing the associated phylogenetic diversity has progressed very well. A total of 260 of the 299 genera of KNP have been sampled and sequenced for *rbcL*, including all trees and shrubs and 60% of grasses. The results have been prepared in the following draft manuscript (Duthoit *et al*., see Annex 15).

4.3.5 South African Conservation Scientists network enhanced

The high level of publicity generated by the two high-profile publications produced from the project have gone a long way to enhancing the network of South African Conservation Scientists. Presentation of these results, both at national conferences and within the national press, has led to a great level of interest in the project amongst local scientists in South Africa, as well as the greater public.

Collaboration with a number of institutions has been enhanced as a result of the DNA barcoding publication. Continued research activities, and the corresponding publicity created as a result, have strengthened links with SANPARKS, and the UJ team now has an excellent working relationship with the SANPARKS authorities, and regularly reports research findings to them at the annual KNP network meetings and during visits to the KNP by UJ staff.

The training activities and research visits between Imperial College London, RBG Kew, UCT and UJ have strengthened the network between these four institutions. With the ever-increasing research on DNA barcoding, UJ have put themselves at the forefront of major DNA barcoding initiatives:

1) The Consortium for the Barcode of Life (CBOL) conference in New York was attended by Dr Michelle van der Bank and Mr Olivier Maurin, with a presentation of the research undertaken in KNP; as a result UJ have been invited to participate in CBOL's Leading Labs Network;

2) Tree-BOL, a newly established worldwide research project designed to barcode all of the 100,000 tree species of the world, has coordinated research into regional working groups, of

which Dr van der Bank is the 'Regional Working Group Co-Chairperson for Africa', and is thus responsible for coordinating and reporting upon all of the DNA barcoding activities which are being undertaken in Africa.

The publication of a special issue of *Molecular Phylogenetics and Evolution*, arising from the SAAB/SASSB VII meeting in South Africa in January 2008, highlights the close working relationship between Conservation Scientists working in South Africa and wide range of research topics that these Scientists now collaborate on and communicate to each other. A total of 12 papers have been fully edited and formally accepted by Drs Savolainen, Dreyer (Univ. Stellenbosch) and Verboom (UCT); they will be published as a theme issue early next year.

4.4 Project standard measures and publications

All of the project standard measures are quantified in Annex 4, and full details of all publications pertaining to the project are provided in Annex 4 too.

The overall project has had two papers published in high-profile journals (Forest, Grenyer *et al*., 2007, *Nature*; Lahaye *et al*., 2008, *PNAS*) and, given the large amount of publicity these publications received and the close working relationship between the host institutions involved and the South African governing institutions (i.e. SANPARKS), these publications have the potential to cause a sizeable impact in the fields of conservation assessments and DNA barcoding. Whilst a considerable degree of the collection and initial analysis of the PD *Nature* paper was initiated during the original project, the editors and reviewers of Nature asked us to undertake many new analyses undertaken during this post-project, and we have enclosed as an annex the paper and its supplementary material (Annex 11, 11a, 11b and 11c).

There has been considerable previous research undertaken in the search for a universal barcode for the angiosperms, and our results represent the largest survey undertaken and have identified the gene *matK* as a viable barcode. Although this information is still embargoed, following our *PNAS* publication, *matK* has been subsequently chosen as one of the two regions by TreeBOL and the Plant Working Group of CBOL.

4.5 Technical and Scientific achievements and co-operation

The two principal areas of scientific research conducted by this project have been DNA barcoding, and conservation assessments. All of the publications resulting from this project are detailed in Annex 4 and pdf's are provided as annexes (Annexes 10-16).

The extensive DNA resources available within South Africa as a result of this project and the original Darwin project, from which this arose, have been utilised to conduct the single largest study on DNA barcoding, with the flora of the KNP now recognised as a case study in this field. Along with the enhancement of existing partnerships, several new partnerships have also been developed, for example the Tree-BOL initiative. Baseline data have been, and continue to provide data for conservation actions within South Africa, through the PD analyses and assessment of extinction risks.

To date four publications have been published in peer-reviewed journals from the project's research, and four further papers submitted for publication. Two of the resulting publications, published in *PNAS* and *Nature* (see abstracts below).

A further publication on DNA barcoding tested the suitability of other DNA regions as putatitive DNA barcodes (Lahaye R, Savolainen V, Duthoit S, Maurin O, Van der Bank M. 2008. A test of *psbK-psbI* and *atpF-atpH* as potential plant DNA barcodes using the flora of the KNP as a model system (South Africa). *Nature Preceedings:hd1:10101/npre.2008.1896.1*). Provided as annex 17.

PAPERS PUBLISHED, EXAMPLE OF ABSTRACTS

- DNA barcoding the floras of biodiversity hotspots
Proceedings of the National Academy of Sciences, USA 105: 2923-2928

Renaud Lahaye, Michelle van der Bank, Diego Bogarin, Jorge Warner, Franco Pupulin, Guillaume Gigot, Olivier Maurin, Sylvie Duthoit, Timothy G. Barraclough, and Vincent Savolainen

DNA barcoding is a technique in which species identification is performed by using DNA sequences from a small fragment of the genome, with the aim of contributing to a wide range of ecological and conservation studies in which traditional taxonomic identification is not practical. DNA barcoding is well established in animals, but there is not yet any universally accepted barcode for plants. Here, we undertook intensive field collections in two biodiversity hotspots (Mesoamerica and southern Africa). Using >1,600 samples, we compared eight potential barcodes. Going beyond previous plant studies, we assessed to what extent a ''DNA barcoding gap'' is present between intra- and interspecific variations, using multiple accessions per species. Given its adequate rate of variation, easy amplification, and alignment, we identified a portion of the plastid *matK* gene as a universal DNA barcode for flowering plants. Critically, we further demonstrate the applicability of DNA barcoding for biodiversity inventories. In addition, analyzing>1,000 species of Mesoamerican orchids, DNA barcoding with *matK* alone reveals cryptic species and proves useful in identifying species listed in Convention on International Trade of Endangered Species (CITES) appendixes.

- ***Preserving the evolutionary potential of floras in biodiversity hotspots***
Nature 445: 757-760

Félix Forest, Richard Grenyer, Mathieu Rouget, T. Jonathan Davies, Richard M. Cowling, Daniel P. Faith, Andrew Balmford, John C. Manning, Serban Proches, Michelle van der Bank, Gail Reeves, Terry A. J. Hedderson & Vincent Savolainen

One of the biggest challenges for conservation biology is to provide conservation planners with ways to prioritize effort. Much attention has been focused on biodiversity hotspots. However, the conservation of evolutionary process is now also acknowledged as a priority in the face of global change. Phylogenetic diversity (PD) is a biodiversity index that measures the length of evolutionary pathways that connect a given set of taxa. PD therefore identifies sets of taxa that maximize the accumulation of 'feature diversity'. Recent studies, however, concluded that taxon richness is a good surrogate for PD. Here we show taxon richness to be decoupled from PD, using a biome-wide phylogenetic analysis of the flora of an undisputed biodiversity hotspot—the Cape of South Africa. We demonstrate that this decoupling has real-world importance for conservation planning. Finally, using a database of medicinal and economic plant use, we demonstrate that PD protection is the best strategy for preserving feature diversity in the Cape. We should be able to use PD to identify those key regions that maximize future options, both for the continuing evolution of life on Earth and for the benefit of society.

-A test of psbK-psbI and atpF-atpH as potential plant DNA barcodes using the flora of the KNP as a model system (South Africa).
Nature Preceedings:hd1:10101/npre.2008.1896.1 2008

Lahaye R, Savolainen V, Duthoit S, Maurin O, Van der Bank M.

No abstract but full text available at: http://precedings.nature.com/documents/1896/version/1

EXAMPLE OF PAPER IN PREPARATION

- ***The Meaning of Extinction Risk in Plants***

To be submitted to Nature

T. Jonathan Davies, Gideon F. Smith, Yolande Steenkamp, John C. Manning, Peter Goldblatt, Timothy G. Barraclough, Jonathan van Alphen Stahl, Richard M. Cowling, Craig Hilton-Taylor, & Vincent Savolainen

It is estimated that up to 37% of species may be lost by 2050 if current extinction trends continue. In the most well known groups of organisms (i.e. mammals, birds and amphibians), the risk of extinction is far greater within evolutionary distinct and species-poor lineages. In addition, species traits, such as body size, fecundity, and geographic range are important predictors of vulnerability. Although plants are the basis for life on Earth, very little is known about the patterns and drivers of their extinction. Here we show that the risk of extinction is unevenly distributed across the plant tree-of-life, and that geographic location, rather than species attributes, is the primary determinant of risk. Assembling comprehensive phylogenetic, distribution and conservation assessment data for the two best-known national floras, namely the United Kingdom (UK) and South Africa, we demonstrate that young, rapidly diversifying lineages are most at threat. Reproductive biology and ecological strategy are generally poor indicators of vulnerability, and specific traits, such as pollination, affect taxa differentially. These results are in sharp contrast to patterns found across animals, where biology can be key to survival in the face of increasing environmental change. In plants, species traits affect the distribution of taxa, influencing community structure, but extinction risk appears to be largely determined by the environment in which a species ends up. Individual species traits may therefore play only a minor role in community resilience. Our results are of critical importance for reducing rates of biodiversity loss in plants. First, to maximise the preservation of the tree-of-life for plants, current criteria for assessing threat are insufficient. Second, because threat is concentrated geographically in 'extinction hotspots', conservation strategy should differ between plants and animals, with the former, we should focus on areas rather than taxa, whereas selected taxon-based approaches may be more appropriate for the latter.

PAPER IN PRESS (Although not directly linked to our post-project, this paper is part of the Special Issue on the Cape in Mol. Phyl. Evol. Edited by the network and coauthored by project partners, and also uses PD)

- ***Dissecting the plant–insect diversity relationship in the Cape***
Molecular Phylogenetics and Evolution, *In Press, Corrected Proof*, *Available online October 2008*

Şerban Procheş, Félix Forest, Ruan Veldtman, Steven L. Chown, Richard M. Cowling, Steven D. Johnson, David M. Richardson, Vincent Savolainen

It has been argued that insect diversity in the Cape is disproportionately low, considering the unusually high plant diversity in this region. Recent studies have shown that this is not the case, but the precise mechanisms linking plant diversity and insect diversity in the Cape are still poorly understood. Here we use a dated genus-level phylogenetic tree of the Cape plants to assess how plant phylogenetic diversity compares with taxonomic diversity at various levels in predicting insect diversity. We find that plant phylogenetic diversity (PD) is a better predictor of insect species diversity that plant species diversity, but the number of plant genera is overall as good a predictor as PD, and much easier to calculate. The relationship is strongest between biomes, suggesting that the relationship between plant diversity and insect diversity is to a large extent indirect, both variables being driven by the same abiotic factors and possibly by common diversification, immigration and extinction histories. However, a direct relationship between plant diversity and insect diversity can be detected at fine scales, at least within certain biomes. Diversity accumulation curves also indicate that the way plant phylogenetic diversity and the number of plant genera increase over spatial scales is most similar to that for insect species; plant species show a greater increase at large spatial scales due to high numbers of local endemics.

4.6 Capacity building

The training of host country researchers by UK project partners has represented the greatest component of capacity building in the project. Several training activities (see 4.3.2) have built the capacity for research of staff and students in the host country and provide them with the tools required to continue research on biodiversity and conservation of the South African flora.

For DNA banking, Ms Gopal, DNA bank manager at SANBI Kirstenbosch received training in the techniques required to run a DNA bank. Initially there was a visit by Ms Kapinos (RBG Kew) to SANBI Kirstenbosch in March 2007, followed by Ms Gopal visiting the molecular systematics laboratory at RBG Kew (July 2007) to receive in-house training in DNA extraction techniques from Kew's DNA bank managers, Ms Kapinos and Mr Csiba. The training included various DNA extractions and purification methods, preparation of chemical solutions required for DNA extractions, DNA Bank database and documentation issues, DNA quality assessment, and Health and Safety issues. Ms Gopal is the third DNA bank manager employed at SANBI Kirstenbosch; the original DNA bank manger, Ms Balele, received equivalent training at RBG Kew and trained one of the lab's intern students (Ms Roussouw), who subsequently was appointed in her place when she left SANBI to take up a position with the South African Institute for Aquatic Biodiversity (SAIAB). Ms Roussouw left the post to undertake an MSc course at UCT. Each of the three bank managers has provided DNA extraction training to undergraduate students during an annual molecular biology course held at SANBI Kirstenbosch.

For DNA barcoding, training was organised as an intensive one-week course in 'DNA Barcoding: a practical guide' held at UJ's molecular laboratory and run by Dr Powell, Dr van der Bank and several members of the staff working there (Olivier Maurin, Cynthia Motsi and Jerminah Moeaha). All students were awarded an overall percentage based upon the performance in the lab during the week in addition to their final exam. These marks contributed towards their BSc Honours degree.

In accordance with Darwin Initiative reporting guidelines, the capacity of the UK lead institution to be an effective project partner is reported on: RBG Kew has increased its capacity to be effective project partner through the experiences gained over the lifetime of this project. For example, the evaluation forms introduced to assess the training courses provide an invaluable source of feedback, and ensure that future courses will better meet the demands of the attendees.

4.7 Sustainability and Legacy

The Royal Society/South African NRF grant awarded to continue working on the KNP will guarantee that the project's activities will not cease upon the cessation of the project, and will expand upon the data already compiled by the original and post-projects.

In addition, Dr van der Bank's key position as African coordinator in the Tree-BOL project will also provide assurance that the current project's activities will be sustained. By initiating and leading the African campaign, UJ is bringing together scientists from all over Africa, and plays a major role in helping to build scientific capacity in Africa. This will be achieved by providing training to young scientists from selected institutions in molecular techniques at UJ. The ultimate goal for us would be to establish a network of African scientists and institutes working in the field of DNA barcoding that will allow us to be in a position to assume a leading role in international scientific campaigns.

DNA barcoding of all tree species of Africa has already started. SANBI is one of the first organisations to commit and join this *African campaign*. With its preserved plant specimen records kept in its three Herbaria (Pretoria, Cape Town and Durban) totaling nearly two million specimens, the Institute is in a good position to pinpoint the occurrence of trees in their natural habitat, and to re-collect material from precise localities if required. Researchers from SANBI will therefore assist in the collection of the estimated 1700 trees native to southern Africa. These samples will be deposited in the DNA Bank at UJ to facilitate their curation, linking them to herbarium voucher specimens deposited at both JRAU and SANBI (PRE). The project will also assist SANBI to improve its electronic information base on South and southern African trees, and facilitate the expansion of its National Plant Collecting Programme. The project will also enable the transfer of laboratory and other barcoding skills to participating staff members.

The first Tree-BOL workshop for all interested participants took place on the 9-10 October 2008 at UJ. Representatives from eight African countries, which include South Africa, Namibia, Zimbabwe, Mozambique, Kenya, Nigeria, Ethiopia, Mauritius and Benin, attended the workshop. The aim is to identify key species to target as priority, based on their conservation and trade status and also to set up partnerships between the different institutions.

The various training activities that have taken place in both the UK and South Africa have ensured that there will not be a shortage of suitably trained people available to undertake research related to the project's activities.

As a significant signal of the support which one of the South African host institutions has for the project, UJ is planning to submit a proposal to the South African NRF for the molecular systematics laboratory headed by Dr van der Bank to be recognised as a Centre of Excellence in DNA barcoding. The envisaged centre will be associated with SANBI, and both Prof IC Burger (Dean: Faculty of Science) and Prof A Habib (Deputy Vice-Chancellor Research, Innovation & Advancement) from UJ have approved the proposal. The proposal has also been discussed by Dr van der Walt (UJ) and Mr Sowazi (Grant Director, Centres of Excellence & SARchl Programmes) during a visit to UJ on Friday, 26 September 2008.

5 Lessons learned, dissemination and communication

5.1. Lessons

There are several valuable lessons that have been learned from the experiences of this project, and we believe that project partners in both the host country and the UK are now more capable of dealing with the demands of such a project.

PD and phylogeny-based extinction analyses are difficult. We had difficulties filling the post of extinction analyst. First Dr Jonathan Davies took a job in the US and then Jonathan van Alphen Stahl quit the job to start medical studies. In the future we would make sure we have a good 'reservoir' of applicants, for example from those we trained throughout the project.

Following a meeting with the two UCT MSc students it was found that the bursaries given by the project were not sufficient to cover living costs, and were less than other bursaries that were awarded. In the future, we would therefore increase these bursaries.

Some conservation assessments have not yet been published. In particular the paper by Forest on the Gouritz area has been on hold for a long time and is still only at an early stage; and we are unsure about how to enforce commitment to publish. In the future we may need to draw written agreements with project collaborators for deliverables such as publications needing to be submitted by a certain date.

5.2. Dissemination and communications

A lot about dissemination has been described above. We can add here that the dissemination of information relating to project achievements has certainly been to both a scientific and wider public audience, both in the host country and internationally. Several presentations have been given at high-profile national and international conferences by partners involved in the project, ensuring that the scientific community has been made aware of this Darwin Initiative project:

- Evolution meeting, New Zealand (June 2007)

Savolainen, V. Phylogenetic Diversity at the Meso-Scale in South Africa: South Africa's Conservation Conundrum

- Annual Meeting of the Society of Conservation Biology, South Africa (July 2007)

Davies, T. J. Phylogenetic Diversity at the Meso-Scale in South Africa South Africa's Conservation Conundrum

- Second International Barcode Conference (of the Consortium for the Barcode of Life), Taipei (September 2007)

Van der Bank, M. A DNA barcode for all trees and shrubs of the Kruger National Park (KNP)

- 6th Scientific Network, Kruger National Park, South Africa (April 2008)

Van der Bank M, Lahaye R, Maurin O, Duthoit S, Savolainen V. A DNA barcode for all trees and shrubs of the Kruger National Park (KNP).

Van der Bank M, Lahaye R, Maurin O. A molecular phylogeny of the Acacia's of the Kruger National Park (KNP).

- Tree-BOL 2008, The New York Botanical Garden, USA (01-02 May 2008)
Van der Bank M, Jayeola A. Regional diversity, resources, funding and planning – AFRICA.

- Dendrological Society of South Africa (26 July 2008)
Van der Bank M. A DNA barcode for all trees and shrubs of the Kruger National Park (KNP)

Scientific papers that have been published or submitted by project partners are available in Annex 4, along with pdf's annexed (Annexes 10-16).

Dissemination will be ongoing after the project has ended, as future research publications of data gathered during the project will be published, and presentations of the research outcomes will continue.

5.1 Darwin identity

All of the publications resulting from the project (see Annex 4) have acknowledged the role of the Darwin Initiative, and all publicity generated by the project has made reference to the role of the Darwin Initiative in the success of the project.

Several presentations have been given at high-profile national and international conferences by people involved in the project (see above), and each of these included the Darwin Initiative logo and acknowledged the support it provided.

A calendar produced by UJ, with images of the flora of the KNP, was accompanied by a leaflet detailing the DNA barcoding project and several hundreds copies were distributed to institutions worldwide. Both the calendar and the leaflet acknowledged the support of the Darwin Initiative.

6 Monitoring and evaluation

The overall purpose of the project was extended from that of the original project, to maximise the potential of the data gathered during that project. The Logical Framework Approach to project management was utilised for the duration of the project, with monitoring carried out in accordance with the five areas described. Regular phone and email contact between project partners allowed for frequent monitoring, evaluation and updating. In addition to this, the workshops that were organised, and spread over the project, allowed the project partners to meet and provided an excellent forum for monitoring the progress of the project against the Logframe.

Monitoring of the financial situation was principally carried out between the Finance Department of RBG Kew (Mr G. Sarkis), Mrs Sandy Smuts of UCT and Mr Tinus Fourie of UJ.

6.1 Actions taken in response to annual report reviews

The reviews provided in response to annual reports were always discussed between project partners, and other collaborators where relevant, and the response to issues raised were agreed jointly between project partners. All of the issues raised in response to the annual reports have been addressed over the course of the project, and these can be summarised as follows:

1. *This work is interesting and important, and has been leading to high profile publications. However, one of these publications (Forest et al. 2007) was submitted from the previous Darwin grant, while much of the data in the second (Lahaye et al. 2008) has been obtained from Mesoamerican orchids. I would like to see an estimate of how much of this research results directly from, and would not have been achievable without, this Darwin funding. For example, the Boatwright et al. paper that was submitted in the first year of this project*

acknowledges the Darwin Initiative for "DNA aliquots supplied" but it is not clear how many aliquots were 'supplied' and how many were generated by the authors?

RESPONSE: As explained in this report, a lot of the work for the Forest *et al*. paper was undertaken during this post-project (i.e. revisions and additional analyses requested by *Nature*). The paper by Lahaye *et al*. combines Mesoamerican orchids and a multi-gene dataset from the KNP. All analyses were supervised by Dr Savolainen during workshops funded by the Darwin Initiative. The paper was written by Drs Savolainen, Lahaye and van der Bank during a workshop funded by the Darwin Initiative. Thus, the Darwin Initiative was key to the successful completion of this high profile publication. When Dr Savolainen went to UJ, he not only participated in the activities listed in the report but also worked with the students on their various phylogeny projects, assisting with the analyses and the writing up of the papers. Some of these projects also benefited from the DNA material banked through this project. Hence we consider that the paper by Boatwright is also a by-product, in part, of this project (pdf provided, annex 12).

2. *The appointment of an Extinction Risk Analyst appears to have been dogged by bad luck – are there any lessons to be learned here?*

RESPONSE: There is a shortage of trained phylogeny-based extinction analysts in South Africa. A training course on the use of PD in conservation was run during the first year of the project at UCT, which was attended and well received by 18 participants. The publication by Forest, Grenyer *et al.* in *Nature* also served to highlight the importance and use of this method in conservation assessments, and raised awareness of the use of PD as a valuable tool for conservation. In addition, a two-day training course on 'Extinction Risk Analysis' was given to 23 participants (staff and post-graduate students) at SANBI Pretoria, and also raised awareness of the usefulness of the methodology, and contributed, although moderately, to filling the potential shortage in people trained in such techniques.

3. *Would it be possible to see more details of student […] how the two post-docs mentioned were funded and what involvement they have in the Darwin project […] mentions a PhD student based at Kew who would be working at the "plant-pollinator interface"; I am not sure who this student is, and what involvement they have had in the project – could this also be clarified?*

RESPONSE: Some details on students are provided in Section 4.3.2 under 'training'. The PhD student based at RBG Kew is Mr Jan Schnitzler, supervised by Dr Savolainen, and his involvement with the project has been to produce some phylogenetic hypotheses for several genera (e.g. *Babiana* and *Moraea*, the latter being used in the extinction analysis paper by Davies, Savolainen *et al*.). The two post-docs were hosted and funded by Dr van der Bank's Molecular Systematics Laboratory at UJ, and worked on DNA barcoding and PD analysis of the KNP, both integral components of this Darwin Initiative post-project.

4. *While person-to-person contact between the UK and the host country is one of the objectives of this grant, as we become increasingly aware of green issues such as air-miles, can it be demonstrated that all trips were necessary and could not have been replaced by videoconference, email, etc?*

RESPONSE: We totally agree that air travel should be limited in a period of climatic change and we are aware of our responsibilities to the environment. As such, trips between the respective countries have only been conducted when necessary, and all trips are described in this report. Video-conferencing facilities are not available at the partner institutes. Email has been used extensively, but of course training activities had to involve flying partners to South Africa. The writing up of the DNA barcoding paper, which was published in *PNAS*, has been the product of intensive brainstorming. This could not have been achieved easily by email, and especially that all sorts of analyses were necessary as well discussing with various members of CBOL – hence Dr Lahaye came to London for some of his work on this paper.

7 Finance and administration

7.1 Project expenditure

	Budget	2006/7	2007/8	2008/9 (April-August)	Total	Variance
Rents, rates, heating, lighting, cleaning, overheads						
Office costs e.g. postage, telephone, stationery						
Staff (Extinction Risk Analyst; DI Project Officer)						
Travel & Subsistence						
Printing						
Conference & Seminars						
Others (MSc & Hons bursaries; Audit fees; DNA consumables)						
Capital items (laptop)						
TOTAL						

7.2 Additional funds or in-kind contributions secured

Additional funds secured during the course of the post-project, which ensure that the research will continue, include the Royal Society/South African National Research Foundation grant 'Regional patterns of biodiversity and conservation in South Africa: the flora of the Kruger National Park as a case study'; this grant is worth £228,451 (to be paid to Dr Savolainen at Imperial) + ZAR 1,593,642 (to be paid to Dr van der Bank at UJ) over 5 years.

The South African NRF (Thuthuka grant) and UJ provided some additional funding for the DNA barcoding of the KNP, including contribution to fieldwork costs and supporting two postdocs (a two-year post for Dr R Lahaye, and a one-year post for Dr S Duthoit) dedicated to this project. UJ also purchased (March 2006) an automated DNA sequencer for the DNA barcoding of the KNP part of the project, worth ca. £60K.

UCT contributed to fieldwork costs and also waived its 10% overheads as matched funding (£1100).

Matched funding arrangements included salary costs for six members of staff at RBG Kew, and nine in South Africa for the percentage of their time dedicated to the project (see table in the application).

7.3 Value of DI funding

The project represents excellent value for money for a number of reasons, and through the support of the Darwin Initiative the image and international awareness of the two South African partner institutions as centres for scientific research has been enhanced considerably.

The scientific publicity created by the project has been considerable, most notably as a result of the publications in *PNAS* (Lahaye *et al.*) and *Nature* (Forest, Grenyer *et al*.). These publications highlighted the role of the South African institutions in the research and referenced the Darwin

Initiative itself; equivalent publicity would be costly. The patent on 'DNA Barcoding of Plants' that we applied for with UJ may bring significant income in the future to these partners.

The development of the molecular systematics laboratory at UJ into one of CBOL's leading DNA barcoding laboratories and a proposed centre of excellence for DNA barcoding highlights the value of the DI funding. Whilst UJ had already received funding from the South African NRF for DNA barcoding work, the assistance of this project cannot be understated in propelling UJ to the forefront of DNA barcoding activities in Africa, particularly following the publication of the *PNAS* paper.

Annex 1 Report of progress and achievements against final project logframe for the life of the project

Project summary	Measurable Indicators	Progress and Achievements April 2007 - March 2008	Actions required/planned for next period
Goal: *To draw on expertise relevant to biodiversity from within the United Kingdom to work with local partners in countries rich in biodiversity but constrained in resources to achieve* *The conservation of biological diversity,* *The sustainable use of its components, and* *The fair and equitable sharing of the benefits arising out of the utilisation of genetic resources*			*(do not fill not applicable)*
Purpose (i) co-ordinate research; (ii) calculate extinction risks; (iii) transfer knowledge to in-country scientists, students and conservationists with regard to the use of phylogenetic data and DNA barcodes; (iv) integrate the human dimension; (v) publish concerted conservation actions	1. Research and training activities in partnership with academic and governmental sectors increase 2. Awareness of biodiversity issues increase among students and young scientists 3. In-country CBD strategy and monitoring of 2010 targets take into account post-project outputs and outcomes	Research and training activities have been highly successful and in high demand throughout the duration of the project. The publicity generated by both the *Nature* paper last year, and *PNAS* paper this year have served to greatly increase awareness amongst the scientific and general community of the biodiversity issues South Africa are faced with. SANBI is step-by-step integrating our results in its CBD strategy.	
Output 1. DNA barcoding	500 DNA barcodes produced	Almost 800 DNA barcodes have been produced for South African taxa.	
Activity 1. DNA extractions and sequencing for DNA barcoding and for hotspots-wide surveys		DNA extraction and sequencing facility at UJ is expanding and being used by other groups, both within UJ and from national	

		institutions (e.g. SANBI Pretoria and Kirstenbosch).
Output 2. Training	20 training-weeks, 2 MSc, 3 Hons, 2 post-docs (total 89 month-person)	51 training weeks (excluding MSc and Hons); 2 MSc due to complete at UCT by project end; 2 Hons completed at UJ and 1 scheduled for completion by July 2008; 2 post-docs, Drs Sylvie Duthoit and Renaud Lahaye trained at UJ.
Activity 2. Course		A total of 4 training courses were run by UK project partners in South Africa (UCT: 'Use of phylogenetic diversity in conservation'; UJ: 'DNA barcoding: a practical guide' and 'Scientific grant writing'; SANBI Pretoria: 'Extinction Risk Analysis')
Output 3. Dissemination	5 papers submitted/2 newsletters circulated	2 newsletters circulated (Kew Scientist/UJ magazine); 7 papers published, in press or in prep (Forest, Grenyer *et al*., Lahaye *et al*. (x2), Duthoit et al., Davies et al., Boatwright *et al*., Proches *et al*.)
Activity 3. Data compilations; assessing extinction risks		Data compilation complete for all South African genera and extinction risks analyses complete for the three target regions (Cape, Gouritz and KNP).
Output 4. Conservation assessments	3 assessments published	1 assessment published for the Cape (Forest *et al*. *Nature*); 2 preliminary assessments made, for Gouritz (Forest *et al*. *in prep*.) and KNP (Duthoit *et al*. *in prep*.)
Activity 4. PD analyses		PD analyses completed for the Cape (Forest et al. & Davies et al.); Cape and Gouritz results presented at the Evolution meeting in June 2007 and Annual Meeting of the Society of Conservation Biology in July 2007. PD analyses for Gouritz and KNP completed and manuscripts in preparation.
Output 5. South African Conservation Scientists network enhanced	>15 staff working together	Savolainen, Powell, van der Bank, Hedderson, Lahaye, Duthoit, Forest, Manning, Tolley, Gopal, Proches, Boatwright, Verboom, Smith, Bogarin, Kapinos, Maurin, Smith, Dreyer, van Alphen Stahl, Faith, Grenyer etc.
Activity 5. Workshops		Initial project workshop in July 2006 at SASSB conference, and final project workshop at UJ on 28-29 July 2008. Other workshops for monitoring and publication writing (*PNAS*).

Annex 2 Project's final logframe, including criteria and indicators

Project summary	Measurable indicators	Means of verification	Important assumptions
Goal: *To draw on expertise relevant to biodiversity from within the United Kingdom to work with local partners in countries rich in biodiversity but poor in resources to achieve the conservation of biological diversity, the sustainable use of its components, and the fair and equitable sharing of the benefits arising out of the utilisation of genetic resources*			
Purpose *(i) co-ordinate research; (ii) calculate extinctions risks; (iii) transfer knowledge to in-country scientists, students and conservationists with regard to the use of phylogenetic data and DNA barcodes; (iv) integrate the human dimension; (v) publish concerted conservation actions*	*1. Research and training activities in partnership with academic and governmental sectors increase* *2. Awareness of biodiversity issues increase among students and young scientists* *3. In country CBD strategy and monitoring of 2010 targets take into account post-project outputs & outcomes*	*1. Joint supervision and research documents and correspondence between SANBI, UJ, UCT & Kew* *2. Records of requests to undertake Hons/MSc, participate in projects, and attend courses by students and young scientists* *3. Conservation & CBD documents updated*	*Strategies developed throughout the post-project are of high quality and in demand by wider scientific and nature conservation authorities* *Joint programme of activities has proven useful and partnership continues* *SANBI's statutory mission continues to be supported by Government*
Outputs			

1.DNA Barcoding *2.Training* *3.Dissemination* *4.Conservation assessments* *5. South African Conservation Scientists network enhanced*	*1. 500 DNA barcodes produced* *2. 20 training-weeks, 2 MSc, 3 Hons, 2 postdocs (total 89 month-person)* *3. 5 papers submitted/2 newsletters circulated* *4. 3 assessments published* *5. >15 staff working together*	*1. DNA sequences available in GenBank* *2. Attendees lists/diplomas* *3. Manuscripts available, correspondence with editors/publishers* *4. Reports available* *5. Meeting reports available*	*There is a broad interest from staff and students for training and networking in biodiversity and conservation* *Material produced is of good quality & accepted for publication* *Collecting permits continue to be issued by KNP*
Activities	*Activity Milestones (Summary of Project Implementation Timetable)*		
1.DNA extractions and sequencing for DNA barcoding and for hotspots-wide surveys *2.Data compilations; assessing extinction risks* *3.PD analyses* *4.Workshops* *5.Course*	*Months 1-12:* *Data compilations (IUCN, phylogenies) and extinction risks analyses start (08/06); training course analyses at UCT (01/07); 2 MSc and 2 Hons research projects start (02/07); presentation of results at conference (06/07); one paper submitted (08/07);* *Months 13-24:* *Darwin Initiative officer start (09/07); 250 DNA barcodes produced (11/07); 2 Hons completed (11/07); Workshop at SASSB VII ((01/08); 2 Hons start (02/08); Extinction risks analyses completed (02/08); 1 paper submitted and 1 press release (03/08); 250 additional DNA barcodes produced; 3 additional papers submitted (06/08); Workshop (07/08); Conservation assessments completed and reports produced (07/08).*		

Annex 3 Project contribution to Articles under the CBD

Project Contribution to Articles under the Convention on Biological Diversity

Article No./Title	Project %	Article Description
6. General Measures for Conservation & Sustainable Use	5	Develop national strategies that integrate conservation and sustainable use.
7. Identification and Monitoring	10	Identify and monitor components of biological diversity, particularly those requiring urgent conservation; identify processes and activities that have adverse effects; maintain and organise relevant data.
8. In-situ Conservation	5	Establish systems of protected areas with guidelines for selection and management; regulate biological resources, promote protection of habitats; manage areas adjacent to protected areas; restore degraded ecosystems and recovery of threatened species; control risks associated with organisms modified by biotechnology; control spread of alien species; ensure compatibility between sustainable use of resources and their conservation; protect traditional lifestyles and knowledge on biological resources.
9. Ex-situ Conservation		Adopt ex-situ measures to conserve and research components of biological diversity, preferably in country of origin; facilitate recovery of threatened species; regulate and manage collection of biological resources.
10. Sustainable Use of Components of Biological Diversity	5	Integrate conservation and sustainable use in national decisions; protect sustainable customary uses; support local populations to implement remedial actions; encourage co-operation between governments and the private sector.
11. Incentive Measures		Establish economically and socially sound incentives to conserve and promote sustainable use of biological diversity.
12. Research and Training	45	Establish programmes for scientific and technical education in identification, conservation and sustainable use of biodiversity components; promote research contributing to the conservation and sustainable use of biological diversity, particularly in developing countries (in accordance with SBSTTA recommendations).
13. Public Education and Awareness	5	Promote understanding of the importance of measures to conserve biological diversity and propagate these measures through the media; cooperate with other states and organisations in developing awareness programmes.
14. Impact Assessment and Minimizing Adverse Impacts		Introduce EIAs of appropriate projects and allow public participation; take into account environmental consequences of policies; exchange information on impacts beyond State boundaries and work to reduce hazards; promote emergency responses to hazards; examine mechanisms for re-dress of international damage.
15. Access to Genetic Resources	10	Whilst governments control access to their genetic resources they should also facilitate access of environmentally sound uses on mutually agreed terms; scientific research based on a country's genetic resources should ensure sharing in a fair

Article No./Title	Project %	Article Description
		and equitable way of results and benefits.
16. Access to and Transfer of Technology	10	Countries shall ensure access to technologies relevant to conservation and sustainable use of biodiversity under fair and most favourable terms to the source countries (subject to patents and intellectual property rights) and ensure the private sector facilitates such assess and joint development of technologies.
17. Exchange of Information	5	Countries shall facilitate information exchange and repatriation including technical scientific and socio-economic research, information on training and surveying programmes and local knowledge
19. Bio-safety Protocol		Countries shall take legislative, administrative or policy measures to provide for the effective participation in biotechnological research activities and to ensure all practicable measures to promote and advance priority access on a fair and equitable basis, especially where they provide the genetic resources for such research.
Other Contribution		Smaller contributions (eg of 5%) or less should be summed and included here.
Total %	100%	Check % = total 100

Annex 4 Standard Measures

Code	Description	Totals (plus additional detail as required)
Training Measures		
1a	Number of people to submit PhD thesis	N/A
1b	Number of PhD qualifications obtained	N/A
2	Number of Masters qualifications obtained	2 to be submitted (Alastair Potts and Matthew Britton, UCT); **Target 2: TARGET NEARLY MET, i.e. pending submission and award of MSc**
3	Number of other qualifications obtained	N/A
4a	Number of undergraduate students receiving training	19 (3 honours degrees: Phip Moolman, Genevieve Thompson and Anneli van Rooyen; 16 honours students attended the UJ DNA barcoding training course); **Target was 3 undergraduates (Hons) for 10 months per year: TARGET EXCEEDED**
4b	Number of training weeks provided to undergraduate students	34 months (3 Hons students for 10 months each; 16 weeks at UJ DNA barcoding training course); **Target was 30 months: TARGET EXCEEDED**
4c	Number of postgraduate students receiving training (not 1-3 above)	21 (14 at UCT course, 2 at UJ grant writing course, 5 at SANBI Pretoria course); **Target 20 was students for 1 week course at UCT**: **TARGET EXCEEDED**
4d	Number of training weeks for postgraduate students	21 weeks (14 at UCT, 2 at UJ, 5 at SANBI Pretoria); **Target was 20 weeks: TARGET EXCEEDED**
5	Number of people receiving other forms of long-term (>1yr) training not leading to formal qualification(ie not categories 1-4 above)	2 >1yr, 2<1yr (Dr Mathieu Rouget 4 months at SANBI Pretoria, Mr Jonathan van Alphen Stahl 6 months at UCT, Drs Renaud Lahaye and Sylvie Duthoit 36 months at UJ); **Target was 1 postdoc for 18 months at SANBI: TARGET EXCEEDED**
6a	Number of people receiving other forms of short-term education/training (ie not categories 1-5 above)	29 (Dr Martyn Powell, DI project officer from October 2007, 13 months; Ms Keshni Gopal, 2 weeks at RBG Kew; 27 researchers attended the various training courses held in South Africa); **Target was 1 DI Project Officer (10 months): TARGET EXCEEDED**

Code	Description	Totals (plus additional detail as required)
6b	Number of training weeks not leading to formal qualification	68 weeks (52 weeks DI project officer, 2 weeks at RBG Kew for Ms Gopal, 4 weeks UCT training course, 5 weeks UJ DNA barcoding/grant writing course, 5 weeks SANBI extinction risk course); **Target was 40 weeks: TARGET EXCEEDED**
7	Number of types of training materials produced for use by host country(s)	3 (UCT 'PD in conservation' course booklet, UJ 'DNA barcoding' course booklet; UJ 'Grant writing' course booklet); **Target was 1 training material produced: TARGET EXCEEDED**
Research Measures		
8	Number of weeks spent by UK project staff on project work in host country(s)	15 (5 Savolainen; 4 Powell; 3 Kapinos; 2 Grenyer; 1 Forest);**Target was 8 weeks spent by UK staff in RSA: TARGET EXCEEDED**
9	Number of species/habitat management plans (or action plans) produced for Governments, public authorities or other implementing agencies in the host country (s)	3 produced (Cape, Gouritz and KNP); **Target was 3 species management plans produced (Cape, Gouritz, KNP): TARGET NEARLY MET (i.e. pending final submission as scientific papers)**
10	Number of formal documents produced to assist work related to species identification, classification and recording.	N/A
11a	Number of papers published or accepted for publication in peer reviewed journals	3 published (Forest *et al.*, *Nature*; Boatwright *et al*., *Syst. Bot*.; Lahaye *et al*., *PNAS*); 1 in press in *Mol. Phyl. Evol*. issue (Proches *et al*.); **Target was 5 peer-reviewed papers: TARGET NOT MET YET, although it will be exceeded when 3 papers in preparation (Davies *et al*., Duthoit *et al*., and Forest *et al*.) are submitted in 2009**
11b	Number of papers published or accepted for publication elsewhere	1 (Lahaye *et al*., *Nature Preceedings*); **ADDITIONAL OUTPUT**
12a	Number of computer-based databases established (containing species/generic information) and handed over to host country	2 (UJ's DNA bank and DNA barcoding databases);**Target was 1 DNA barcoding computer database: TARGET EXCEEDED**
12b	Number of computer-based databases enhanced (containing species/genetic information) and handed over to host country	1 (SANBI's DNA bank database); **Target was 1 DNA bank computer database enhanced: TARGET MET**

Code	Description	Totals (plus additional detail as required)
13a	Number of species reference collections established and handed over to host country(s)	1 (786 barcodes and 260 *rbcL* sequences); **Target was 1 species reference collection (500 barcodes + 200 *rbcL*): TARGET EXCEEDED**
13b	Number of species reference collections enhanced and handed over to host country(s)	3 (Compton, KNP, UJ herbaria); **Target was 2 species reference collections (Compton and KNP herbaria): TARGET EXCEEDED**
Dissemination Measures		
14a	Number of conferences/seminars/workshops organised to present/disseminate findings from Darwin project work	2 project workshops organised (SASSB conference July 2006; final project workshop July 2008); **Target 2 workshops (Darwin projects and final workshop): TARGET MET (although workshop between Darwin projects was not possible)**
14b	Number of conferences/seminars/ workshops attended at which findings from Darwin project work will be presented/ disseminated.	6 (Evolution meeting, New Zealand, June 2007 [special PD symposium]; Annual Meeting of the Society of Conservation Biology, South Africa, July 2007; Second International Barcode Conference [of CBOL], Taipei, September 2007; 6th Scientific Network, Kruger National Park, South Africa, April 2008; Tree-BOL 2008, The New York Botanical Garden, USA, May 2008; Dendrological Society of South Africa, 26 July 2008); **Target was 1 conference attended (Evolution meeting): TARGET EXCEEDED**
15a	Number of national press releases or publicity articles in host country(s)	>10 (e.g. see http://www.uj.ac.za/PlantMolecularSystematicsLaboratory/Researchprojects/TreeBOLAfrica/Pressrelease/tabid/13946/Default.aspx); **Target was 1 press release in host country: TARGET EXCEEDED**
15b	Number of local press releases or publicity articles in host country(s)	1 (Kruger Park Times article);**Targetwas 1 local press release: TARGET MET**
15c	Number of national press releases or publicity articles in UK	>50 (e.g. Defra, BBC, Nature, RBG Kew, Imperial College); **Target was 1 press release in UK: TARGET EXCEEDED**
15d	Number of local press releases or publicity articles in UK	N/A
16a	Number of issues of newsletters produced in the	2 (Kew Scientist and UJ news magazine); **Target was 2**

Code	Description	Totals (plus additional detail as required)
	host country(s)	**newsletters (SANBI and Kew Scientist): TARGET MET**
16b	Estimated circulation of each newsletter in the host country(s)	N/A
16c	Estimated circulation of each newsletter in the UK	N/A
17a	Number of dissemination networks established	N/A
17b	Number of dissemination networks enhanced or extended	2 (CBOL, Tree-BOL); **Target was 1 network enhanced (PD/barcoding): TARGET EXCEEDED**
18a	Number of national TV programmes/features in host country(s)	1 (Dr van der Bank: SABC News, February 2008); **ADDITIONAL OUTPUT**
18b	Number of national TV programme/features in the UK	N/A
18c	Number of local TV programme/features in host country	N/A
18d	Number of local TV programme features in the UK	N/A
19a	Number of national radio interviews/features in host country(s)	3 (Dr van der Bank: RSG, morning talks, 2008; Radio Pretoria, 2008; RSG - Eco-Forum, 2008); **ADDITIONAL OUTPUT**
19b	Number of national radio interviews/features in the UK	N/A
19c	Number of local radio interviews/features in host country (s)	N/A
19d	Number of local radio interviews/features in the UK	N/A
Physical Measures		
20	Estimated value (£s) of physical assets handed over to host country(s)	1 laptop (£800)
21	Number of permanent educational/training/research facilities or organisation established	N/A
22	Number of permanent field plots established	N/A
23	Value of additional resources raised for project	Ca. £500k
Other Measures used by the project and not currently including in DI standard measures		
	N/A	

Publications

Type * (eg journals, manual, CDs)	Detail (title, author, year)	Publishers (name, city)	Available from (eg contact address, website)	Cost £
MAIN PUBLICATIONS OF THIS POST-PROJECT				
Proceedings of the National Academy of Sciences, USA 105: 2923-2928	DNA barcoding the floras of biodiversity hotspots Renaud Lahaye, Michelle van der Bank, Diego Bogarin, Jorge Warner, Franco Pupulin, Guillaume Gigot, Olivier Maurin, Sylvie Duthoit, Timothy G. Barraclough, and Vincent Savolainen 2008	National Academy of Sciences, USA	http://www.pnas.org	USD 250 research article reprint rights
PAPER STARTED DURING ORIGINAL PROJECT AND FINISHED DURING POST-PROJECT				
Nature 445: 757-760	Preserving the evolutionary potential of floras in biodiversity hotspots Félix Forest *, Richard Grenyer *, Mathieu Rouget, T. Jonathan Davies, Richard M. Cowling, Daniel P. Faith, Andrew Balmford, John C. Manning, Şerban Procheş, Michelle van der Bank, Gail Reeves, Terry A. J. Hedderson, and Vincent Savolainen 2007	Nature Publishing Group (NPG)	www.nature.com	20
ADDITIONAL PUBLICATIONS PARTLY DUE TO THIS POST-PROJECT				
Systematic Botany 33 (1): 133-147	Systematic Position of the Anomalous Genus *Cadia* and the Phylogeny of the Tribe Podalyrieae (Fabaceae)	American Society of Plant Taxonomists, Wyoming, USA		

	James S. Boatwright, Vincent Savolainen, Ben-Erik van Wyk, Anne Lise Schutte-Vlok, Félix Forest, and Michelle van der Bank 2008			
Molecular Phylogenetics and Evolution In press	Dissecting the plant–insect diversity relationship in the Cape Şerban Procheş, Félix Forest, Ruan Veldtman, Steven L. Chown, Richard M. Cowling, Steven D. Johnson, David M. Richardson, and Vincent Savolainen	Elsevier, St. Louis, USA	http://www.elsevier.com	

Another 3 publications are in preparation and will be submitted shortly (pdf are annexed):

- Davies *et al*. ***The meaning of extinction risks in plants***. Manuscript fully drafted (annex 14).
- Duthoit *et al*. ***Trees and shrubs taxon richness, phylogenetic diversity and environmental correlates in the Kruger National Park (South Africa): towards an improvement of conservation actions?*** Manuscript fully drafted (annexes 15, 15a, 15b). Will be submitted in 2009 as soon a paper on functional ecology of the KNP is written (this work by a black PhD student at UJ uses the same phylogenetic tree and we are concerned somebody will do these same analyses before the PhD student if we release the KNP phylogeny too early on with the paper by Duthoit *et al*.)
- Forest *et al*. ***Evaluating biodiversity patterns in the Little Karoo of South Africa: phylogenetic diversity and ecosystem status***. Early draft enclosed (annex 16).

Annex 5 Darwin Contacts

Ref No	EIDP013
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Role within Darwin Project	Project leader
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