Darwin Initiative

Annual Report

1. Darwin Project Information

Project Ref. Number	14-001
Project Title	Conservation and Monitoring of Meso-American
	Orchids
Country(ies)	Costa Rica
UK Contractor	Royal Botanic Gardens, Kew (RBG Kew)
Partner Organisation(s)	Lankester Botanical Garden (LBG), University of Costa Rica (UCR)
	Centro de Investigación en Biología Celular y Molecular (CIBCM, UCR)
	Sistema Nacional de Áreas de Conservación (SINAC)
	Ministerio de Ambiente y Energía (MINAE)
Darwin Grant Value	£ 151,900 (Total)
Start/End dates	1 June 2005 to 31 May 2008
Reporting period (1 Apr	1 June 2005 to 31 March 2006
200x to 31 Mar 200y) and annual report number (1,2,3)	Report 1
Project website	http://www.jardinbotanicolankester.org
Author(s), date	Vincent Savolainen (RBG Kew, UK), Guillaume Gigot
	(RBG Kew, UK), Jorge Warner (LBG, Costa Rica)
	and Diego Bogarin (LBG, Costa Rica), 25/04/2006

2. Project Background

Costa Rica is one of the richest tropical countries in terms of biodiversity, and especially for orchids with about 1,300 species within just 51,100 km². Lots of conservation efforts have been undertaken by the Costa Rican government to protect a significant portion of its territory, however the CBD's 2010 targets and the Global Strategy for Plant Conservation (GSPC) have brought up new challenges for the Costa Rican scientific authorities. RBG Kew and LBG have been collaborating on orchid ecological and systematic research for several years and as a natural progression, this project aims to develop new expertise in Costa Rica for biodiversity research and conservation of Meso-American orchids.

3. **Project Purpose and Outputs**

The project purpose includes: (a) Provide measures of biological diversity and establish long-term monitoring sites; (b) Develop the necessary strategies, policies

and material transfer agreements (MTAs) for conservation and sustainable use of orchids (including use of genetic material, propagation and potential commercialisation); (c) Undertake a pilot study on DNA barcoding for conservation and trade surveillance; (d) Achieve high standards for research and training in orchid biology and link with global efforts to build the orchid tree-of-life; (e) Increase awareness in societal, academic, and politic sectors of the necessity of conserving the epiphytic flora; (f) Help implement CBD's GSPC and the orchid action plan of the IUCN/SSC.

Project outputs include several staff and students trained in DNA Barcoding, phylogenetics and conservation of orchids, conservation assessments for monitoring orchid diversity, several publications and species/DNA reference collections & DNA barcodes.

4. Progress

This project was initiated in June 2005 and this is the first reporting period, which therefore corresponds to the first ten months of the project.

4.1 Summary of progress

During this first period LBG has been CITES registered as a scientific institution entitled to the exemption provided by Article VII, paragraph 6 of the Convention, intensive fieldwork has allowed for LBG's orchids collections to be significantly increased and DNA barcoding of those species has now started at Kew. Progress has been monitored according to five main activities: 1) Training, 2) Conservation assessments, 3) Collecting, 4) Policies and strategies for orchid conservation, 5) Research & education networking.

A Summary of progress over the last year, against the logical framework, is provided in Annex 1.

Mr Guillaume Gigot has been appointed as Darwin Initiative Project Officer at Kew, and Mr Diego Bogarin has been appointed as Darwin CBD Implementation Officer at LBG/UCR in Costa Rica; both are well-qualified fellows and also are fluent in English and Spanish.

4.1.2 Training

From 7th to 14th of July 2005, the main project partner in the host country, Prof. Jorge Warner, visited RBG Kew to launch the project. During this period, the Memorandum of Cooperation was drafted with the help of Dr. Noel McGough (Head of Kew's Convention And Policies Section), Phyllida Walsh (Legal Consultant) and Kate Davis (Kew's CBD officer). Material Transfer Agreements (MTAs) were also discussed with M. Groves (Kew's CITES officer), especially with regard to exchanging orchid material under CITES regulations. Financial arrangements were set up with Des Bennett and George Sarkis (Kew's Finance Department). J. Warner also met with Dr. Phillip Cribb (Assistant keeper of the Kew Herbarium), to talk about the organisation of the 3rd International Orchid Conservation Congress (III IOCC), which will take place in San Jose, Costa Rica, in 2007. One seminar, attended by about 30 persons, was given by J. Warner at the Jodrell Laboratory to present LBG's research and conservation activities (11th of July 2005).

During this trip J. Warner also visited Dr. Phil Seaton from the Orchid Specialist Group (OSG) to discuss the use of Red Listing, the participation of LBG in the OSG and to review the performance of the OSG's Meso-American group.

From March to May 2006, UK project partners visited Costa Rica to discuss the monitoring strategy, set up the training course and participate with fieldwork activities. The Darwin Initiative project officer, Guillaume Gigot, went to Costa Rica

from March 16th to May 12th, project partner Martyn Powell from March 24th to April 26th, and project leader Dr. Vincent Savolainen from April 4th to April 24th.

Despite the fact that the application stated that 15 students would be trained during the course of this year, totalling an equivalent of 30 weeks, ten undergraduate students has been trained at LBG and 41 training weeks were achieved (of which six weeks were in April).

During the period 16-31 March, G. Gigot visited LBG and UCR to organise the training aspects of the project. From 20-28 March 2006, a workshop was organised by J. Warner and G. Gigot to discuss the training activities, including Phylogenetics and DNA Barcoding for conservation, with Prof. F. Albertazzi from CIBCM-UCR and M. Powell from Kew. It was decided to provide a one week practical course on "Phylogenetics and DNA Barcoding for conservation", which was subsequently attended by six students from 04-07 April 2006 (totalling 6 weeks training). Details of this course are reported in section *4.2 (Difficulties and future work plan/Already done in April 2006*).

Four undergraduate students from UCR have been trained during this reporting period. For their part-time research projects, Anabel Quiros and Jose-Daniel Zuñiga, are now working at LBG, since February 2006 and November 2005 respectively. They monitor the databases of the garden, including all the new collections for the project. A. Quiros is in charge of the living collection database and J.-D. Zuñiga is working on the voucher collection (totalling 15 weeks training). Gustavo Rojas and Adam Karremans have been working part time at LBG since June 2005, on the coordination of different collections of the garden (living collection, spirit vouchers and pollinia collection, totalling 20 weeks training). Those four students have also received training in orchid taxonomy and conservation from Prof. F. Pupulin, Prof. R. Dressler and D. Bogarin.

4.1.3 Conservation assessments

All of the necessary permits for collecting material were obtained in order to carry out the fieldwork (see 4.1.4 *Policies and strategies*).

Monitoring strategy and Red Listing

From 20th of March to 5th of April, six project partners (J. Warner, D. Bogarin, F. Pupulin, V. Savolainen, G. Gigot and M. Powell) discussed the possibilities for the long-term monitoring strategy, and it has been decided that the IUCN Red Listing criteria will be used to work on orchid species. This represents a relevant and practical tool to work with populations of epiphytic plants in tropical forests. All of the databases from LBG (Living Collection, Spirit Collection and Electronic Field Books) will be very useful and well valorised, combined with other sources (Herbarium Collection from UCR (USJ), Museo Nacional Costa Rica (CR) and Instituto Nacional de Biodiversidad (INB)) to complete the Red List application. Red Listing of orchid species will start in the three sites of Tapanti National Park, Coco Island National Park and the Monteverde area (see below).

Tapanti National Park

Fieldwork collections and data from several Costa Rican herbarium databases (USJ, CR, INB, including database and living collection of LBG) have been used and enhanced as the basis for the new inventory of the orchid species of Tapanti National Park, with a total of 190 species recorded. This site has been visited five times, and D. Bogarin, F. Pupulin, J. Warner and G. Gigot have collected 62 samples.

Monteverde

On 8-9 March 2006, J. Warner visited the Monteverde Cloud Forest Reserve to meet with a representative from the Tropical Science Centre, to establish contact and confirm the permits for the collecting and monitoring of orchid species within the reserve, of which 404 species are currently listed in LBG database.

However, it has subsequently been decided that the Alberto Brenes Reserve, owned by UCR, in the area of Monteverde, is a more viable study site. This reserve is less well studied and has already been visited on one occasion by D. Bogarin, and 113 samples were collected. As a result, the list of species has been improved by the addition of four new species (total of 210 species), highlighting its potential as an area of great importance to orchid taxonomy. The practicality of working in this UCR reserve, coupled with the discovery of new species, make it a good candidate for a long-term monitoring site.

4.1.4 Policies and strategies

As explained above, following application by Prof. J. Warner, LBG is now the first CITES registered institute in Costa Rica, since March 2006 (CITES number CR-001).

For the Darwin Initiative project to become an official project of UCR, it needed to be approved by the "Comisión Institucional de Biodiversidad" from UCR; this was achieved in May 2005.

In order to obtain all of the permits required for the project, considerable time has been spent working with the relevant Costa Rican authorities by J. Warner and D. Bogarin. This involved interactions with the three institutions, MINAE, SINAC and UCR, who regulate the three levels of permits required: namely (1) conservation and research, (2) collecting and (3) research and access. Consequently, LBG staff members now have collecting permits for the entire country. Concerning the UNESCO World Heritage Site of Coco Island, J. Warner has established a good working relationship with MINAE and this national park is now a proper site of collecting and monitoring for LBG.

In addition, UCR - the largest scientific institute in Costa Rica - has identified this Darwin Initiative project as a model for all research projects that are carried out in accordance with all national and international legislation.

The Memorandum of Cooperation (MoC) for this project was signed on 15 January 2006 by Dr. Yamileth González Garcia (Rector of UCR) and Prof. Sir Peter Crane (Director of RBG Kew).

4.1.5 Publications and publicity

A general presentation of the project has been published in issue 28 of Kew Scientist (October 2005). This is an international newsletter reporting on news from the Living Collections, the Herbarium and the Laboratories at Kew and Wakehurst Place, available online at www.kew.org/kewscientist (see Annex 2, page 5).

Project partners from LBG have organised the 3rd International Orchid Conservation Congress for March 2007, to be hold in San Jose, Costa Rica. The congress is already well publicised and will be attended by over 200 participants. Darwin Initiative staff are involved (J. Warner as General Coordinator, F. Pupulin as Scientific Coordinator) and the project will be presented at this meeting. Project partners have discussed that the printing of the conference proceedings will be supported by our Darwin funds.

The LBG website has now been designed and launched, it will incorporate more details of this project in the near future.

A press release in Costa Rica was supposed to be published this year. In February 2006, an article giving a general presentation of the project has been accepted by the journal *Epidendrum* edited by UCR. This paper is in press in the issue 27 of *Epidendrum* but it has not been published yet due to financial difficulties at the editorial office of UCR.

4.1.6 Collections and DNA Barcoding

A total of 14 fieldtrips have been conducted in Costa Rica since the beginning of the project and 1,231 orchid samples have been collected by D. Bogarin, F. Pupulin and R. Dressler. At LBG the orchid collections have been increased with 100 live specimens, 100 duplicated silica-dried samples for DNA extraction, and 100 vouchers have been done, for a total of 100 species. The living collection of LBG now totals over 16,000 individuals, which represents substantial sampling for DNA extraction and barcoding.

At Kew, samples from Kew's DNA Bank have been used to start the barcoding work on orchids from Costa Rica and 100 DNA sequences have been produced (representing 44 species). Work has focused on five different regions of the plastid genome (*trnH-psbA*, *matK*, *accD*, *rpoB* and *rpoC1*). Initially, 50 sequences have been produced for *trnH-psbA* and we are now producing sequences on the four other regions proposed to be good candidates for the barcoding of land plants by the Plant Working Group of the Consortium for the Barcoding of Life (CBOL).

The work on digitisation of Kew herbarium type specimens has started and following the training of G. Gigot in digitisation techniques 50 scans have already been done.

4.2 Difficulties and future work plan

Several activities, including the visit to Costa Rica by UK partners, fieldwork, training and transfer of material between LBG and RBG Kew, have been delayed principally due to administrative reasons and took place in April 2005 (just after this reporting period ending March 2005). It was indeed difficult to start the aforementioned activities before the signature of the Memorandum of Cooperation, which was signed on 15 January 2006. The project has become a model of application of MTAs and national and international legislations for UCR, consequently the administrative process took longer than expected.

In addition, a further regulatory body, Comisión Nacional para la Gestión de la Biodiversidad (CONAGEBIO), created in 2004 and part of MINAE, is now involved in the issue of Research and Access permits. This has necessitated increased discussion and coordination between LBG, UCR and MINAE, with the inevitable consequence of an increase in the time required for the application of those permits.

Already done in April 2006

A one week practical training course, given by F. Albertazzi, G. Gigot and M. Powell, on "Phylogenetics and DNA Barcoding for conservation" has been attended by six students from CIBCM at UCR, from 04-07 April 2006. On a practical level they have learned the basic molecular biology techniques from DNA extraction through to DNA sequencing. In addition, they followed the basic introduction to DNA sequence analyses and a seminar on the concept of DNA barcoding. As part of this training session two one-hour seminars given by project leader V. Savolainen have been attended by 30 students and researchers of UCR, on the 6th and 7th of April 2006. The first seminar introduced the international programme 'Assembling the Tree of Life'. The second one presented more in-depth uses of phylogenetic trees, especially with regard to understanding speciation processes such as sympatric speciation in palms on an Australian oceanic island. In addition, mains partners have discussed training contents and three topics are now highlighted: DNA Barcoding, Red Listing and GIS.

A visit to Coco Island has been carried out from 11-21 April 2006, by J. Warner, D. Bogarin, V. Savolainen and G. Gigot, to collect multiple samples of the five orchid

species from the island, including an attempt to clarify the status of the rare species *Epidendrum jimenezii*. During this fieldwork, 100 samples dedicated to DNA extraction and molecular population study, have been collected and 38 living samples will increase the LBG collection. Coco Island represents an ideal location to establish a long-term monitoring plot to study the changes in the vegetation and for Red Listing and we discussed the possibility of creating a one hectare plot as an objective for the next visit on the island.

In May 2006, a seminar on "Phylogenetics and DNA Barcoding for conservation" will be given by G. Gigot at LBG and attended by approximately 8 LBG staff and students. A focus list of species from Tapanti NP will be established in order to evaluate the Red Listing possibilities is this National Park; the aim will be to submit five species to the Red List.

In July 2006, talks relating to DNA barcoding will be given by V. Savolainen and G. Gigot at the Sixth Conference of the Southern African Society for Systematic Biology.

From August-October 2006, D. Bogarin will visit RBG Kew to receive training in molecular biology techniques and herbarium digitisation.

Before the end of 2006, one press release in Costa Rica (La Nación).

In March 2007, the III IOCC will take place in San Jose, Costa Rica.

5. Actions taken in response to previous reviews (if applicable)

Not applicable

6. Partnerships

Following several years of excellent collaboration between LBG and RBG Kew the Darwin Initiative project has further strengthened the link between these two institutions and as such the project has profited from this solid base.

The six principal partners (J. Warner, D. Bogarin and F. Pupulin from LBG, and V. Savolainen, G. Gigot and M. Powell from RBG Kew) interact routinely to coordinate their activities and are in regular communication in both languages.

The two Darwin Initiative project officers hired this year have integrated well into the dynamics of the project and have very well enhanced the interaction and collaboration between the two institutions.

As a direct consequence of the project, interaction between Costa Rican partners, within UCR has been enhanced, particularly between LBG and CIBCM, especially Prof. F. Albertazzi.

7. Impact and Sustainability

All the efforts of LBG during this year have been rewarded by the CITES registration, the first in Costa Rica, which will facilitate transfer of material for any future project between LBG and any other overseas institutes. This provides a solid foundation for attracting future research projects and activities for the conservation of biodiversity.

The regular communication between J. Warner and all different components of MINAE concerning the permits applications have enabled LBG to establish a privileged relationship with these government authorities, which will be beneficial for future activities of the garden.

The Darwin Initiative project manager in Costa Rica, D. Bogarin, was hired in August 2005, but has already been involved in LBG activities since January 2003. He is an excellent taxonomist, specialised in orchids, and displays a willingness to learn new techniques. As such, he will receive training in herbarium digitisation and molecular biology techniques during a forthcoming visit to RBG Kew, which has already been discussed and planned for 2006. He is an ideal candidate to apply for a permanent position at LBG, where new skills in DNA sequencing, DNA Barcoding and DNA banking will be valorised to develop further activities and projects for LBG. It will also allow the development of interdisciplinary activities within UCR, for example, through the collaboration with CIBCM.

8. Outputs, Outcomes and Dissemination

Most of our targets have been met as detailed in Table 1. However, we partially achieved the training output (15 students and 30 training weeks) by totalling 35 weeks with 4 students from UCR, due to the fact that the visit of UK partners in Costa Rica has been delayed for logistic (trip to Coco Island) and administrative reasons as reported above. It was impossible to coordinate with UCR and to organise lectures and training course with more students before April 2006.

Concerning the LBG orchids collections, the target of the next reporting period (*At least 200 orchid species samples collected*) has already been reached: at least 300 species have been collected and identified by D. Bogarin, in only eight month (from August 2005 to March 2006). This strong effort of collecting represents an important potential of work for DNA barcoding and data available for monitoring the diversity of orchid in the host country.

Code No.	Description	Year 1 Total	Year 2 Total	Year 3 Total	Year 4 Total	TOTAL
4C	4 students trained in Taxonomy and database monitoring at LBG	4				
4B	One week training per student, total 35 weeks	35				
5	1 CBD Project Officer hired and trained for	1				
6A	Two staff to set MoC and MTAs at Kew	2				
6B	1 week workshop	1				
8	Gigot 2 weeks & Powell 1 week in March 2006 (total 3 weeks)	3				
15	Item in Kew Scientist in October 2005	1				
20	2 computer laptop + 1 ext. hard disk	£2,200				

Table 1. Project Outputs (According to Standard Output Measures)

Type *	Detail	Publishers	Available from	Cost £
(e.g. journals, manual, CDs)	(title, author, year)	(name, city)	(e.g. contact address, website)	
Kew scientist item listed				
under press releases				

Table 2: Publications

9. Project Expenditure

10. Monitoring, Evaluation and Lessons

The main partners of both institutes (J. Warner, D. Bogarin, G. Gigot and V. Savolainen) exchange all the necessary information required for monitoring the project on a monthly basis via email and telephone contacts. The project website requires improvement to be useful in monitoring and evaluation of the project. The aim will be to update regularly a description of the progress and to have LBG and project databases updated available online.