Darwin Initiative Annual Report

Darwin Project Information

Project Ref Number	14-001
Project Title	Conservation and Monitoring of Meso-American Orchids
Country(ies)	Costa Rica
UK Contract Holder Institution	Royal Botanic Gardens, Kew (RBG Kew)
UK Partner Institution(s)	
Host country Partner Institution(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 of Project	1 June 2005 to 31 May 2008
Reporting period (1 Apr 200x to 31 Mar 200y) and annual report number (1,2,3..)	1 April 2006 to 31 March 2007 Annual report 2
Project Leader Name	Vincent Savolainen
Project website	http://www.jardinbotanicolankester.org/ing/project_a.html
Author(s), date	Vincent Savolainen (RBG Kew, UK), Guillaume Gigot (RBG Kew, UK), Jorge Warner (LBG, Costa Rica) and Diego Bogarin (LBG, Costa Rica), April 2007

1. Project Background

Among the richest tropical countries in terms of biodiversity, Costa Rica hosts an extraordinarily diverse orchid flora, with over 1,300 species on a relatively small territory of 51,000 km^2. In spite of the fact that this country has a well-developed network of protected areas, with over 25% of its territory composed of protected forests and reserves, the orchid flora remains under constant threat from factors such as deforestation and illegal trade. 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. Started in June 2005, the project has now obtained all the required permits and LBG has become the first institution in Costa Rica to be CITES-registered for exemption from Article VII. MTAs between the two partner institutions have been set up and are in use. The team is currently working on various activities, including a sustained collecting effort, the development of a DNA barcode for orchids and training courses. In addition, partners have started to work on IUCN-Red List conservation assessments.

2. Project Partnerships

Over the last year, the two main partner institutions (Kew & LBG) showed a solid collaboration based on the regular and frequent interaction of the project leader and in-country coordinator, as well as the project officers who exchanged information weekly/monthly by email or phone. Kew's partners have spent 16 weeks at LBG-UCR and D. Bogarin (CBD implementation officer at LBG) has spent 12 weeks at Kew during the summer of 2006.

Several workshops at LBG, UCR and Kew have further strengthened the partnership, especially regarding the topics of phylogenetics, DNA barcoding and new uses of LBG's database for GIS work and Red List assessment. These workshops have led to new collaborations in Costa Rica, e.g. between LBG and Prof. Francisco Aguilar from the Laboratorio de Geomaticá (UCR) to valorise LBG's geo-referenced data.

The organisation of the third International Orchid Conservation Congress (IOCC) has been an exceptional activity for LBG's staff and this Darwin project. It has been a good opportunity to re-enforce many contacts and to publicise this project, especially with UCR's administrative authority and MINAE. Kew has helped LBG with the organisation of the congress during a visit to Costa Rica in March 2007.

Other Collaborations

The third IOCC had a noticeable impact within the orchid research and conservation community. In total 144 attendees and 23 countries were represented. Various activities were included in the IOCC's program, e.g. a workshop on "Population dynamics applied to orchid conservation", and several meetings of IUCN/SSC Specialist Groups. The congress program is available online (http://www.jardinbotanicolankester.org/ing/congress.html).

For this event, we have contacted three other Darwin projects involved in the use of molecular tools for species identification, forensics and conservation: "Molecular tools for promoting biodiversity in rainforest fragments of Borneo" (Ref. 10-025), "Certification to support conservation of endangered Mexican desert cacti" (Ref. 14-059), and "Building genetic forensic capacity to reduce South Africa's illegal trade" (Ref. 13-018). A joint poster outlining these projects under the hospice of the Darwin Initiative was presented during the IOCC congress (see annex).

Contact has been established with another Darwin Initiative project in Costa Rica and Panama: "Baseline Tools for Management of PN La Amistad" (ref. 15027) and a joint one-day meeting was organised on 26th of October 2006 to discuss common ground, attended by Dr Savolainen (project leader Kew), Mr Gigot (Kew), Mr Bogarin (LBG) and Mr Alex Monro (project leader Natural History Museum).

New collaborations have been established in the host country, e.g. with G. Biamonte from APREFLOFAS (Association for the Preservation of the Wild Flora and Fauna). Dr Biamonte also attended our training course on GIS and Red List assessment in April 2007 at UCR. APREFLOFAS is a private, non-profit, non-governmental organization. It was established in Costa Rica in 1985 to fight for conservation of nature through direct actions supported by research and environmental education programs (www.preserveplanet.org). Currently APREFLOFAS chairs the Costa Rican Members Committee of the World Conservation Union (IUCN).

3. Project progress

3.1 Progress in carrying out project activities

Training

Several training activities took place this year, totalling 12 staff training weeks and 42 student training weeks. This result meets project objectives for this period, in addition to a few seminars and courses dedicated to improving students' and staff skills in conservation assessments.

Six students have attended a one-week training course at UCR on "DNA barcoding and phylogenetics for conservation", from 04-07 April 2006.

At LBG, in May 2006, five undergraduate students from UCR, one PhD student from the University of Mexico and two staff members (F. Pupulin and J. Warner) have attended a one-day seminar on "DNA barcoding for conservation" organised by Kew.

Mr Bogarin, Darwin Initiative project officer in Costa Rica (CBD implementation), visited Kew between 28th of July and 30th of October 2006. As part of his Masters Degree, he has been trained during 12 weeks in different molecular techniques, from DNA extraction to sequencing in the orchid genus *Scaphyglottis*. Mr Bogarin has also been trained in the digitisation techniques used to scan orchid type specimens at the Kew Herbarium.

In August 2006, four staff members, V. Savolainen, G. Gigot, M. Powell and D. Bogarin, attended a Red List assessment course at Kew given by Dr Craig Hilton-Taylor, who is in charge of the Red List at the IUCN.

Four undergraduate students from UCR have been trained during this reporting period (total 36 weeks). They are undergoing part-time internships at LBG and are currently working on databasing and the ‘Epidendra’ e-field guide (www.Epidendra.org). Jose-Daniel Zuñiga, Gustavo Rojas and Adam Kellermans have worked on the various databases (i.e. of herbarium vouchers, pollinia, silica and living collections) and have been trained by F. Pupulin, D. Bogarin and R. Dressler in Taxonomy and Conservation. Daniel Ramírez worked with F. Pupulin on the development of Epidendra, which is now operational at www.Epidendra.org.

<u>Conservation assessments, monitoring strategy and Red List</u>

Mr Steve Bachman, Species Conservation Assessment officer from the GIS section at Kew, has visited LBG and UCR from 29th of March to 14th of April 2007. He took part in the GIS-Red List workshop, organised the GIS-Red List training course in April 2007 and provided expertise on LBG’s databases for GIS work and Red List Assessments. As a result of the workshop, it became clear that LBG’s databases need further improvement before GIS work can start. The different databases –vouchers, pollinia, silica and living collections- need to be linked and geo-referencing has to be done as necessary. For example, LBG’s living collection includes over 17,000 individuals, over 15,000 specimens are registered in the main database, of which approximately 4,000 specimens have been geo-referenced but ca. 3,000 specimens have only textual locality information. At the taxon level (species and sub-species) there are an estimated 1,700 orchid taxa: 190 have fully geo-referenced specimens, 412 have partially geo-referenced specimens and 1,161 have no geo-referenced data. Based on these geo-referenced specimens, 190 preliminary IUCN-Red List assessments were carried out using the software Arcview 3.2 from ESRI and a method developed at Kew (Willis, F., J. Moat, Paton, A. 2003. "Defining a role for herbarium data in Red List assessments: a case study of *Plectranthus* from eastern and southern tropical Africa." Biodiversity and Conservation 12(7): 1537-1552.). As a result, 12 species were evaluated to be in a Threatened category (CR, EN or VU, IUCN 2001). The remaining 178 taxa were known from too few localities, and therefore were evaluated as Data Deficient, thus highlighting the fact that more collecting is required.

Regarding the monitoring field sites, priority has been given to geo-reference specimens from these areas to allow preliminary assessment as described above. Project partners are currently working on available data from Tapanti National Park and A. Brenes Nature Reserve.

Two full Red List conservation assessments have been completed for two Coco Island species, *Epidendrum cocoense* and *E. insulanum*. These assessments have been submitted for comments to Dave Roberts from the IUCN Orchid Specialist Group and the formal submission to IUCN will be completed next year (2007/8).

Coco Island National Park was visited in April 2006 and >100 orchid samples were collected. DNA extraction of those samples has been completed and a population study has started with AFLP primer trials.

3.2 Progress towards Project Outputs

<u>Publications and publicity</u>

In July 2006, our project and results on DNA barcoding of orchids were presented at the Sixth Conference of the Southern African Society for Systematic Biology (SASSB). This was an opportunity to discuss with the members of another Darwin project in which DNA barcoding plays an important part, 'Integrating Evolutionary History and Phylogenetic Measures of Biodiversity into Conservation Planning, South Africa' (EIDPO13, also lead by Dr Savolainen).

The third IOCC was organised in March 2006 and three press releases appeared in the national newspaper La Nacíon. Our DI project was well represented during this congress through three scientific talks and one poster. One large banner presenting the project was designed and exhibited in the main meeting room.

Three scientific papers have been published in the conference proceedings (Lankesteriana vol. 7, No. 1-2): one article on Costa Rican orchid taxonomy, one article presenting the first results on orchid DNA barcoding, and one article on the Epidendra e-field guide (see annexes).

Our Darwin project has supported the printing of a volume of Lankesteriana dedicated to the IOCC and the Darwin Initiative has been acknowledged throughout (the DI logo appears on the opening page and the back cover).

Two additional scientific papers on orchid taxonomy and the description of new species have been accepted for publication in 2007.

The project's orchid e-field guide, Epidendra, designed by project partners F. Pupulin and D. Bogarin, is now available online at: www.Epidendra.org. It uses LBG's taxonomic data and various collections, in addition to Kew's digitised herbarium specimens.

In August 2006, an interview of DI officers G. Gigot and D. Bogarin, was written for the *Orchid Review*, authored by Phil Seaton from the IUCN-Orchid Specialist Group. It will be published in November 2007.

<u>Collections and DNA barcoding</u>

D. Bogarin and F. Pupulin conducted a total of 22 collecting field trips in Costa Rica this year and a total of 847 orchid specimens have been collected, representing more than 400 species. Each monitoring site (Tapanti, A. Brenes reserve and Coco Island) has been visited once. Silica collection has been increased to 720 samples, including samples for 273 species dedicated to DNA barcoding. A specific database dedicated to this collection has been designed and is now in use at LBG.

Regarding the digitisation work, a considerable effort have been made during D. Bogarin's visit to Kew in the summer 2006, when 1,024 scans were produced from Kew's orchid herbarium. It represents an important compilation of taxonomic data from Charles Lankester's collections, and Mesoamerican historic specimens. It includes 236 types and all these specimens provide new access to some rare data for orchid taxonomists. These scans have been duplicated on a hard drive and sent to LBG. This collection of images will be very useful in Costa Rica for all LBG's staff, students or other specialists on request, and many of these scans are to be made available online in Epidendra.

DNA extraction was undertaken for 219 orchid samples from the LBG's collection, plus 100 samples from populations of the five Coco Island species. A DNA matrix of 432 sequences (covering six potential DNA barcodes proposed by the Plant working Group of the Consortium of the Barcoding of Life, see www.kew.org/barcoding), representing 74 taxa and 50 species, has been produced. Genetic analyses have been carried out and the *matK* plastid region has been identified as a good DNA barcode for Mesoamerican orchids. These results have been presented at IOCC and published in the proceedings of the congress (Lankesteriana, 7(1-2)). Efforts are now being made to build a DNA barcode library of the selected region and 104 taxa have been sequenced so far.

3.3 Standard Output Measures

Project Standard Output Measures

Code No.	Description	Year 1 Total	Year 2 Total
Established codes			
4A	No of undergraduate students to receive training	4	10
4B	Number of training weeks to be provided	35	42
4C	No of postgraduate students to receive training	6	4
4D	Number of training weeks to be provided	6	12
5	Number of people to receive at least one year of training.	1 CBD Project Officer trained and hired	
6A	Number of people to receive other forms of education/training	2 staff to set MoC and MTAs at Kew	
6B	Number of training weeks to be provided	1 week workshop	2 weeks workshops
8	Number of weeks to be spent by UK project staff on project work in the host country	3	16
10	Number of individual field guides/manuals to be produced to assist work related to species identification, classification and recording		1 Orchid e-field guide
11A	Number of papers to be published in peer reviewed journals		5
14A	Number of Conferences to be organised		1 IOCC
14B	Number of conferences to be attended		1 SASSB
15A	Number of national press releases in host country		3 (IOCC media coverage)
15 C	Number of national press releases in UK	1 Item in Kew Scientist in October 2005	
20	Asset	1 laptop computer + external hard disk	

Table 1 Publications

Type * (eg journals, manual, CDs)	**Detail (title, author, year)**	**Publishers (name, city)**	**Available from (eg contact address, website)**	**Cost £**
Lankesteriana, 7(1-2): 200-203. 2007	Gigot *et al.* 2007. Finding a suitable DNA barcode for Mesoamerican orchids.	Jardín Botánico Lankester-UCR	PO box 1031-7050 Cartago Costa Rica	Ca. 20
Lankesteriana, 7(1-2): 446-449. 2007	Bogarin & Pupulin 2007. Las orquídeas del Parque Nacional Barra Honda, Guanacaste, Costa Rica.	Jardín Botánico Lankester-UCR	PO box 1031-7050 Cartago Costa Rica	Ca. 20

Lankesteriana, 7(1-2): 178-180. 2007	Pupulin 2007. Epidendra, the botanical database of Jardín Botánico Lankester at the University of Costa Rica.	Jardín Botánico Lankester-UCR	PO box 1031-7050 Cartago Costa Rica	Ca. 20
Willdenowia, July 2007	Pupulin & Bogarin 2007. A second species of Restrepiella (Orchidaceae: Pleurothallidinae)	BGBM Berlin-Dahlem	Freie Universtat Berlin, Koningin-Luise-Str. 6-8 D-14191 Berlin	
Harvard Papers in Botany, 2007	Dressler & Bogarín 2007. A new and bizarre species in the genus Condylago (Orchidaceae: Pleurothallidinae) from Panama.	Harvard University Herbaria	22 Divinity Avenue, Cambridge, Massachusetts 02138, USA	

3.4 Progress towards the project purpose and outcomes

As a result of the pilot study on DNA barcoding, one DNA plastid region (*matK*) has been selected as a suitable barcode for Mesoamerican orchids. An orchid DNA barcode library is now being built at Kew. The digitisation of Kew's orchid specimens has been completed providing access in Costa Rica to essential taxonomic data. Training activities have been strengthened at UCR through new interdisciplinary collaborations and several training courses. Sustained effort has been made to improve LBG's databases and collections in order to develop routine Red List assessments and GIS activities, which will first focus on the protected areas and monitoring sites of this project. Awareness in societal, academic and political sectors of the necessity of conserving the epiphytic orchid flora has been increased through the organization of the third IOCC.

3.5 Progress towards impact on biodiversity, sustainable use or equitable sharing of biodiversity benefits

The knowledge and expertise in orchid ecology and conservation that is being enhanced at LBG has tremendous potential for playing an important role in decision-making processes with regard to protected areas in Costa Rica. As just one example, the recent work on Barra Honda National Park by D. Bogarin, CBD implementation officer in this project, has reinforced the need to protect this unique ecosystem.

During the workshop at UCR on GIS and IUCN-Red List assessment, the *Sample Red List* Index project was also discussed (www.kew.org/gis/projects/srli/index.html). The Sample Red List is an IUCN initiative in response to CBD's 2010 target and global challenge of "significantly reducing the current rate of loss of biodiversity by 2010". This work aims to evaluate changes in status of a random selection of species (1000 of each major group of organisms). Kew is involved in the coordination and development of methodologies for this project. On a total of 1000 species of angiosperms chosen for the Sample Red List, 28 species are Costa Rican orchids. These species will be prioritised for our work on Red List assessment in order to contribute to this important global target of the CBD.

4. Monitoring, evaluation and lessons

To monitor the project, project targets have been included in a 'project-monitoring table', which is updated monthly by the project partners, using regular email and phone contact.

Last year we have been weak with regard to communicating the project to a wider audience. This year, we have made use of several communication means, especially websites, including our re-designed project website, the e-field guide Epidendra and online material for the training course on GIS/Red List. Communication of the project has also been enhanced with the organisation of the third IOCC, with several press releases that have appeared in a national newspaper and the presence of UCR's TV journalists during the congress.

Following our various workshops and courses, Red Listing appeared to be the most appealing methodological approach to monitoring the flora of Costa Rica. Conservation authorities, UCR and LBG all are very excited about the prospect of assessing orchid species locally and globally, and the substantial database of LBG, once updated, will provide a formidable resource towards meeting this goal.

5. Actions taken in response to previous reviews

Requests by the reviewer from the previous annual review have all been carefully considered and actions were detailed in our mid-year report. Further actions have been taken:

Following the reviewer's comments, we have designed a 'Darwin training certificate' and an evaluation form for the second course, which was organised in April 2007 at UCR. These documents were added to the folder distributed to the trainees in addition to a disc compiling the main data and slideshow of the course.

On the subject of Material Transfer Agreements (MTAs), project partners are strictly following the rules written in the project's Memorandum of Collaboration, signed between partner institutions. It includes the description of a Notification of Transfer form, which is used for each transfer of plant material.

5.1 Other comments on progress not covered elsewhere

N/A

6. Sustainability

The privileged relationship established between LBG and Costa Rican government authorities, especially concerning research and collecting permits, are regularly reinforced by our project activities and fieldwork in the whole country. The LBG is regarded as an expert facility for conservation of the epiphyte flora and regularly provides advice to government officials. Once we have completed our biodiversity monitoring and red listing activities this academia/governmental relationship is likely to become even stronger and contribute to a more efficient and practical implementation of the CBD. Our CBD project implementation officer in Costa Rica is also likely to be hired by UCR/LBG after the Darwin funding period.

7. Dissemination

As mentioned above, significant efforts have been made this year to disseminate results. The newly designed project website has been operational for several months now and is linked to Epidendra online, these websites provide access to various pieces of information from our project (e.g. databases). Several short papers have also been published encompassing the three main aspects of the project: (i) DNA barcoding as a new tool for conservation; (ii) better taxonomy and species discoveries; (iii) databases and online resources (see Table 1 above). These results have also been presented at the IOCC and a conference in South Africa. For the next year we plan high-profile papers, e.g. on DNA barcoding with another Darwin project that works on the DNA barcoding of the Kruger Park in South Africa.

8. Project Expenditure

Please expand and complete Table 3.

Annex 1 Report of progress and achievements against Logical Framework for Financial Year: 2006/07

Project summary	Measurable Indicators	Progress and Achievements April 2006 - March 2007	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*		Studies dedicated to protected areas have been improved. Progress has been made on our orchid diversity monitoring strategy and approach to meet CBD targets.	
Purpose To create in Costa Rica a multi-site expert centre for biodiversity research and conservation on meso-American orchids by: (a) establishing long-term monitoring sites for CBD 2010 targets & GSPC, (b) increasing capacity building in 6 overseas biodiversity institutes, (c) developing material transfer agreements and new conservation strategies for the epiphytic orchid flora.	1.Research and training activities in partnership with public and private sectors increase 2.In-country strategy and CBD policies in place; MTA in use; 2010 targets monitored 3.Collections & DNA barcodes used for taxonomy and forensics 4.Awareness of biodiversity issues increase	A DNA region has been identified as a suitable DNA barcode. Digitisation work has been completed. Training activities have been strengthened at UCR. MTAs are in use and several workshops took place on conservation strategy and assessments. Improved databases & collections to develop routine Red List assessment and monitoring activities. An international congress has been successfully organised and widely publicised by LBG.	Increase DNA Barcoding library for Mesoamerican orchids. Continue Red List assessments and improve utility of LBG's database for more GIS work and studies. D. Bogarin will visit Kew during summer 2007.
Output 1. 1.Staff & students trained	1. 2(1) training weeks to 15 students & 8 staff p.a.	16 staff-training weeks and 42 student-training weeks achieved.	
Activity 1.1 Staff training		In August 2006, 4 members of staff have attended a Red List assessment course (4 weeks).	

		In August-September 2006. D. Bogarin from UCR/LBG has been trained in molecular systematics and specimen digitisation at Kew (12 weeks).	
Activity 1.2 Students training in Costa Rica		6 students have attended a 1-week training course at UCR on DNA barcoding and phylogenetics for conservation, in April 2006. 4 students monitor LBG databases, e-field guide & are trained in Taxonomy and Conservation (total 36 weeks)	Continue training at UCR and LBG. 1 student will participate in fieldwork on Coco Island in April 2007 (1 week). 1 week training course in GIS and Red List assessment in April 2007 (15 students)
Output 2. 2.Habitat/spp assessments and monitoring plots	2.Coco & Tapanti plots & orchids assessed	Several collecting field trips have been organised in these sites. LBG's enhanced database is being updated for routine GIS work and IUCN-Red List pre-assessment and full assessment.	
Activity 2.1. Visit and samples collection		Each monitoring sites –Tapanti NP, A. Brenes reserve and Coco Island have been visited once.	Coco Island has been visited in April 2007
Activity 2.2. LBG's database and Red List assessment		S. Bachman from Kew has helped evaluate LBG's database for GIS potential and Red List assessment. 190 Red List pre-assessments have been completed. Two full assessments completed for Coco Island.	Submit to IUCN the two first Red List full assessments. Continue pre-assessments and complete other full assessments.
Output 3. 3.Publications	3. e-field guide + 4 papers accepted	E-field guide has been designed. 4 papers on orchid taxonomy and 1 paper on DNA barcoding have been published or accepted for publication.	
Activity 3.1. E-field guide		LBG's orchid e-field guide, Epidendra, is now online. It uses LBG's databases, in addition to Kew's digitised herbarium specimens.	Increase number of described species included online. Include more of Kew's digitised specimens.
Activity 3.2. Papers		3 papers have been published in IOCC proceedings: 1 on orchid	More papers to be submitted

		taxonomy, 1 on orchid DNA barcoding & 1 on Epidendra e-field guide. 2 additional papers on orchid taxonomy have been accepted for publication in 2007.	
Output 4. 4.Species/DNA reference collections & DNA barcodes established/enhanced	4. DNAs orchid (600), ex situ collection (ca ½ orchid flora) & herbarium available for use	With 22 fieldtrips done this year by F. Pupulin and D. Bogarin, LBG collections have been significantly enhanced and Kew have started to extract DNA from LBG's samples. A substantial digitisation's effort has been made and over 1000 scans from Kew's herbarium have been produced, duplicated and are now available at LBG.	
Activity 4.1. LBG's living and silica collections		22 collecting field trips have been done in the country and a total of 847 specimens have been collected (more than 400 species). Silica collection has been increased to 720 samples, including 273 species samples dedicated to DNA barcoding.	Reach the target of 600 species in the silica collection available for DNA barcoding
Activity 4.2. Orchid DNA collections		At Kew, orchid DNA collection includes 319 samples (220 species).	Continue DNA extraction from LBD silica-dried samples.
Activity 4.3. Orchid DNA barcoding		At Kew, 464 trial barcodes have been produced for over 50 species, from which 104 *matK* sequences have been selected as suitable barcodes.	Continue sequencing in order to build a DNA barcode library for Mesoamerican orchid.
Activity 4.4. Kew's herbarium specimens digitisation		1024 scans produced from Kew 's Herbarium, including 236 types (representing more than 800 species).	
Output 5. 5.Dissemination	5. Conference organised + 3 workshops + 4 press releases	IOCC has been successfully organised in March 2007 and 5 project partners have attended 2 workshops. The main effort of communication has been to publicise the orchid congress where the project was particularly well represented.	
Activity 5.1. Conferences		In March 2006, the third IOCC took place in San Jose, organised by LBG: 3 national press releases in	

		Costa Rica, project represented by 4 talks, 1 poster and 2 banners.	
Activity 1.4 Workshops		In August 2006, D. Bogarin visited Kew and 4 staff took part in a workshop on phylogenetics and DNA barcoding. In March-April 2007, 5 staff took part in a workshop between partners at LBG and UCR on GIS & Red List assessment.	
Activity 5.3. Press release		An interview with both DI officers has been done for the Orchid Review. It will be published in November 2007.	
Output 6. 6.Meso-American orchid network enhanced	6. Network activities increase	Several visits to the partner institutions have been made by project members. In Costa Rica, IOCC has enhanced LBG's status at international level. A workshop in March 2007 has created new collaborations and potential for new projects utilising LBG's taxonomic data.	
Activity 6.1. Project partners dynamic		4 Kew members of staff have visited LBG and UCR in April 2006 and March-April 2007. 1 LBG staff visited Kew for 3 months in 2006.	
Activity 6.2. IOCC impact		Organised by LBG (23 countries were represented). Various activities within orchid research community.	
Activity 6.3. GIS network within UCR		6 members of staff from Kew, LBG and UCR have attended a GIS-Red List workshop and planned future activities and the organisation of a training course	Continue improvement and use of LBG's database for GIS work and Red List assessment
Output 7. 7.New research & education facility at UCR	7. Facility running	In progress. 2 members of staff and 4 students continue the work to link the databases. 1 student working to manage and sort LBG's databases in order to make it valuable for GIS.	

Annex 2 Project's full current logframe

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 benefits arising out of the utilisation of genetic resources			
Purpose To create in Costa Rica a multi-site expert centre for biodiversity research and conservation on meso-American orchids by: (a) establishing long-term monitoring sites for CBD 2010 targets & GSPC, (b) increasing capacity building in 6 overseas biodiversity institutes, (c) developing material transfer agreements and new conservation strategies for the epiphytic orchid flora.	*1.Research and training activities in partnership with public and private sectors increase* *2.In-country strategy and CBD policies in place; MTA in use; 2010 targets monitored* *3.Collections & DNA barcodes used for taxonomy and forensics* 4.Awareness of biodiversity issues increase	*1.Joint supervision and research documents and correspondence between Lankester, UCR, MINAE, NGOs & RBG Kew* *2.MTA, conservation & CBD documents updated @ MINAE* *3.Records of requests and visits to collections* 4.Records of visits & participation by public to conservation activities	*Strategies developed throughout the project are of high quality and are in demand by wider scientific and nature conservation authorities* *Joint programme of activities has proven useful and partnership continues* Public interest in conservation, especially of orchids, continues to be high enough to support in-country biodiversity initiatives & reserves
Outputs *1.Staff & students trained* *2.Habitat/spp assessments and monitoring plots* *3.Publications* *4.Species/DNA reference collections & DNA barcodes established/enhanced* *5.Dissemination* *6.Meso-American orchid network enhanced* 7.New research & education facility @ UCR	*1. 2(1) training weeks to 15 students & 8 staff p.a.* *2.Coco & Tapanti plots & orchids assessed* *3.e-field guide + 4 papers accepted* *4.DNAs orchid (600), ex situ collection (ca 1/2 of orchid flora) & herbarium available for use* *5.Conference organised + 3 workshops + 4 press releases* *6.Network activities increase* 7.Facility running	*1.Attendees lists* *2.Conservation assessments documents* *3.Correspondence* *4.DNAs duplicated according to MTA & online databases @ Lankester* *5. Registration and attendees lists, press* *6.Correspondence & joint documents from partners* *7.Annual reports from Lankester & other partners*	*There is a broad interest from staff, researchers and students for training and networking in orchid biology, biodiversity and conservation, and to attend conference and workshops* *Material produced is of good quality & accepted for publication* *Collecting permits are issued by MINAE* Lankester Garden's statutory mission continues to be supported by UCR
Activities *1.Specific training* *2.Assessing conservation status* *3.Collecting* *4.Setting up policies and strategies for in and ex situ orchid conservation and sustainable use (incl MTA & CBD 2010)* 5.Research & education networking		*Yr1: MoU (07/05); Conserv assessment Tapanti (02/06); Univ training (11/05); 200 orchid spp collected (04/06); web site launched (03/06); staff hired (08/05).* *Yr2: Workshop (02/06); Conserv assessment Coco/Monteverde (03-04/06); Training (11/06); orchid pollination work and re-assess Tapanti (03/07); 200 spp collected (04/07); DNA barcoding (07/06).* Yr3: International Orchid Conservation Congress (03/07); Conservation re-assessments (04/07); training (11/07); Collect 200 spp (03/08); global conservation strategy (02/08); e-field guide (02/08); exhibition (03/08)	

Annex 3 onwards – supplementary material (optional)

Copies of

Darwin Poster

List of attendees for Darwin Course on GIS & Red Listing

Three papers deriving from the project activities

Checklist for submission

	Check
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Molecular tools and DNA Barcoding for Conservation

The Darwin Initiative (DI) for the Survival of Species promotes biodiversity conservation and sustainable use of resources around the world (http://www.darwin.gov.uk). It is funded and administered by the UK Department for Environment, Food and Rural Affairs, (DEFRA). The main goal of the DI is to assist countries rich in biodiversity but poor in resources with the conservation of biological diversity and implementation of the Biodiversity Convention. Projects funded under the DI are collaborative, involving either local institutions or communities in the host country in collaboration with a British institution.

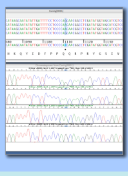
Here we present four DI projects using molecular tools for species identification, forensic use and conservation.

Project in Costa Rica (Ref. 14-001)

Conservation and Monitoring of MesoAmerican orchids

Royal Botanic Gardens, Kew and Lankester Botanical Garden - University of Costa Rica. *Contacts: Mr. J. Warner, Mr. D. Bogarin, Prof. F. Pupulin, Dr. V. Savolainen, Prof. M. Chase, Mr. G. Gigot*

Started in 2005, the project purpose is to record **orchid diversity** and to establish long-term monitoring sites to study Costa Rican orchids.



Main activities:

- to undertake a pilot study on **DNA barcoding** for conservation and trade surveillance
- to provide **research training** in orchid biology, linking with global efforts to build the orchid tree-of-life

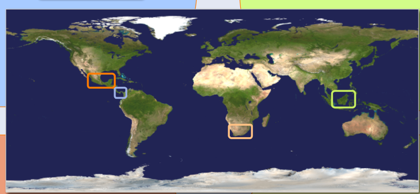
Training course at UCR

DNA barcoding is a diagnostic technique for identifying species using a short DNA sequence from a standardized and agreed-upon region in the genome; such DNA barcode sequences are very short relative to the entire genome and they can be obtained reasonably **quickly** and **cheaply**.

Monitoring site Coco Island

The use of a **standardized identification tool** could provide many potential uses and applications, for example: identification of different life stages (e.g. seeds and seedlings), identification of fragments of plant material, forensics, verification of herbal medicines/foodstuffs, biosecurity and trade in controlled species, inventories and ecological surveys.

http://www.jardinbotanicolankester.org/ing/project_a.html



Project in Borneo (Ref. 10-025)

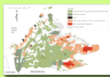
Molecular tools for promoting biodiversity in rainforest fragments of Borneo

University of York, University of Leeds, Natural History Museum, Institute of Tropical Biology and Conservation University Malaysia Sabah, Forest Research Centre Sabah and the Yayasan Sabah.

Contacts: Dr. K. C. Hamer, Prof. M. Maryati and Dr. V. K. Chey

Between 2001 and 2004, this project has focused on land use and environmental changes in **Sabah.** Main objectives were - to provide practical advice on the size and placement of forest patches necessary **to preserve species richness and genetic diversity** - and to assist conservationists in establishing priorities for the conservation of species.

Standard **sequencing** techniques of mitochondrial DNA has been used to study phylogenetic distinctiveness and gene flow/population structure have been estimated **to identify species of high conservation value** and determine their **vulnerability to habitat fragmentation.**



Map showing the study sites. Forest fragments are shown in red.

The team has focused on **butterflies** which are highly diverse with many endemic species on Borneo and are well-known **sensitive indicators** of environmental changes, with the hope that the principles developed by the project will be applicable to a wide range of other taxa.

http://www-users.york.ac.uk/~jkh6/

Project in Mexico (Ref. 14-059)

Certification to support conservation of endangered Mexican desert cacti

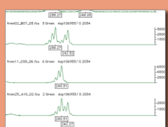
University of Reading, Universidad Autonoma de Querétaro and Universidad Nacional Autónoma de México (UNAM).

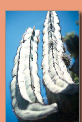
Contacts: Dr. J. A. Hawkins, Dr. R. T. Barcenas Luna and Dr. H. Hernandez

Since September 2005, this project aims to support the conservation, sustainable harvest and use of **Mexican desert cacti** by providing molecular tools which can be used to identify plants to species, to determine their parentage and to locate the populations that they were collected from originally.

These **DNA fingerprinting** and **barcoding tools** are applied here as a **DNA based certification** for the first time to cacti in trade.

The team can match DNA fingerprints of wild plants against those of cultivated plants, to certify genuine nursery-grown stock.





One of the most suitable molecular tools for forensic use is the **microsatellite** method. Plant genomes contain short, repeated sequences called microsatellites or ssrs (simple sequence repeats) that are interspersed within longer more stable sections. The number of times that these short repeated sequences occur varies from individual to individual, and the profile across multiple loci may be specific to a plant and its closest relatives (clonal or sexual offspring), or unique to a population.

Photo credit: R. T. Bárcenas Luna

http://www.uaq.mx/naturales/biologia/ccma/index.html

Project in South Africa (Ref. 13-018)

Building Genetic Forensic Capacity to reduce South Africa's illegal trade

University of Sheffield and University of Kwazulu-Natal.

Contacts: Prof. T. Burke and Dr T. Taylor

The purpose of this project, started in May 2004, is to assist the conservation of wildlife in South Africa through institutional capacity building for **wildlife forensic DNA analysis** (fingerprinting and species identity).

The project initially concentrates on two species currently threatened by **illegal trade**: the Cape Parrot and the Blue Crane.

South African MSc students, trained at Sheffield, use **microsatellites** to genotype individuals and perform paternity analyses for **forensic use**. This technique has been developed to enable claims of captive breeding to be confirmed or refuted in order **to detect illegally caught wild birds**.

This knowledge will be transferred to a **forensic facility** in South Africa. Staff will be trained in genetic forensic analysis, chain of evidence, producing forensic profiles, … Procedures will be drawn up in collaboration with appropriate authorities in South Africa to provide **protocols** and **guidelines**.

The presence of such a high profile technique will raise awareness of the illegal trade and should prove a deterrent, reducing such activities.

http://www.shef.ac.uk/misc/groups/molecol/parrotandcrane.html

This poster has been designed for the 3rd International Orchid Conservation Congress, San Jose, Costa Rica, in March 2007 - Contact: G. Gigot.

Darwin Initiative Project Training Course
GIS and Red List Conservation Assessment

From 09/04/07 to 13/04/07 Laboratorio de Geomática, Universidad de Costa Rica

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FINDING A SUITABLE DNA BARCODE FOR MESOAMERICAN ORCHIDS

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Key Words: DNA barcoding, orchids, Costa Rica, plastid genome, coding region, *trnH-psbA*

Introduction

Recently, DNA barcoding has emerged as an effective tool for species identification. This has the potential for many useful applications in conservation, such as biodiversity inventories, forensics and trade surveillance. It is being developed as an inexpensive and rapid molecular technique using short and standardized DNA sequences for species identification. The core idea of DNA barcoding is based on the fact that short pieces of DNA can be found that vary only to a minor degree within species, such that this variation is much less than between species (Savolainen *et al.* 2005). As proposed by Hebert *et al.* (2003), the DNA barcoding system for animals has been based upon sequence diversity in mitochondrial cytochrome *c* oxidase subunit 1 (COI or *cox1*). However, in land plants, the *cox1* gene has too low a rate of DNA sequence change to be used for species-level discrimination. The plastid genome of plants seems to be a better candidate for DNA barcoding, with enough variation to distinguish species and at the same time less intra- than inter-specific variability (Chase 2005, Cowan 2006). In 2005, Kress *et al.* proposed a non-coding plastid region, the *trnH-psbA* spacer, as a good barcode candidate. The Consortium for the Barcoding of Life (CBOL), via the Plant Working Group, has established another strategy to find a universal DNA barcode for land plants. A subset of six coding regions has been selected and is currently being tested in various plant taxa.

Our study is part of a project funded by the Darwin Initiative for the Survival of Species, which promotes biodiversity conservation and sustainable use of resources around the world (http://www.darwin.gov.uk). This project, based on a partnership between several academic and governmental authorities in Costa Rica with the Royal Botanic Gardens, Kew, in the UK, aims to record orchid diversity, establish long-term monitoring sites and undertake a pilot study on DNA barcoding for conservation and trade surveillance. Although some approaches to identify a DNA barcoding approach for land plants focused on a wide range of species around the world, e.g. the work lead by the Plant Working Group of CBOL, our work concentrates on a limited geographical area, Costa Rica, and a hyper-diverse family of plants, orchids. Costa Rica has one of the richest orchid floras in the world, with over 1300 species on a relatively small territory of 51,000 km^2. In spite of the fact that this country has a well-developed network of protected areas, with over 25% of its territory composed of protected forests and reserves, orchid floras remain under constant threat from factors such as deforestation and illegal trade. Furthermore, orchids are well known to be difficult to identify, particularly when they are sterile. Therefore, the use of a rapid and standardized DNA-based identification tool will be invaluable for many applications in conservation and to enforce international conventions such as the Biodiversity Convention (CBD) and the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). Hence, among other activities of this project, we are currently working on the development of a DNA barcode for Mesoamerican orchids, in particular Costa Rican species.

Nuclear regions such as the internal transcribed spacer of the ribosomal DNA (ITS), although often highly variable in angiosperms, are not a practical option in several groups and show certain functional limitation for DNA barcoding (Kress 2005). Both coding and non-coding plastid regions present various advantages (monomorphy, high copy number and highly diagnostic) and appear to be good candidates. One aim of barcoding is to find a "barcoding gap" between the intra- and inter- specific variation for the proposed regions (Meyer & Paulay 2005). We present here our preliminary results based on the comparison of different DNA plastid regions: the non-coding *trnH-psbA* spacer and five coding regions.

Materials and methods

We used standardized protocols for the PCR amplification and the sequencing available online on the RBG Kew website (http://www.kew.org/barcoding/protocols.html); all DNA samples came from the Kew DNA Bank (http://www.rbgkew.org.uk/data/dnaBank/homepage.html). The sampling covers 74 taxa representing 50 Mesoamerican orchid species and three temperate species as outgroups (from the North Temperate genus *Dactylorhiza*, Orchideae, Orchidoideae). We selected 47 species Costa Rican species and three species from other countries with a more northern distribution in Mesoamerica (from Mexico to Nicaragua).

To evaluate intra-specific variability, eleven of these species had multiple accessions (from two to seven). From the plastid genome, we sequenced the non-coding region *trnH-psbA* and portions of five DNA coding regions that have been put forward by the Plant Working Group of CBOL as potential universal barcodes for land plants, including *accD*, *rpoC1*, *rpoB*, *matK* and *ndhJ*. Altogether, these regions represent an aligned combined matrix of 3698 base pairs (bp) for 74 taxa.

We evaluated the inter- and intra-specific variation from a genetic distance matrix constructed using pair-wise Kimura 2 parameter (K2P) distances. The K2P model was used because it is simple and takes into account variable transition and transversion frequencies. Genetic distance between terminal taxa and their closest sister was used to characterize inter-specific divergence. The two most genetically distant individuals within each species were chosen to represent intra-specific divergence. We compared phylogenetic trees constructed using neighbour joining and parsimony methods. We also combined gene regions to evaluate the potential of a multi-locus barcode.

Results and discussion

Amplification was generally successful with all the regions tested. The only region that presented significant difficulties was *trnH-psbA;* there were alignment problems due to high levels of length variation. The sequence variability within and between species for all gene regions appears to overlap considerably, and, thus, these data do not show any evidence that there is a barcoding gap. Species groupings within neighbour joining and parsimony trees showed no topological differences. At the intra-specific level, the three gene regions that provided the greatest resolution were *matK*, *trnH-psbA* and *rpoB*, grouping over 50% of the eleven species with replicates into monophyletic groups. Among all combinations of regions tested as multi-locus barcodes, a "triplet" of *rpoC1*, *rpoB* and *matK* appeared to provide the best result and grouped all accessions of individuals correctly (Table 1).

Conclusion

As has been found in many plant groups (palms etc.), orchids exhibit low inter-specific sequence divergence, and there is no "barcode gap" between intra- and inter-specific data. However, results from the regions evaluated here show it is possible to

TABLE 1. Number of intra-specific species groupings per gene region from a neighbour joining tree (based on 11 species with replicates).

All regions are coding except for *trnH-psbA*.

Gene regions	Number of species groupings
accD	3 (27.3%)
matK	10 (90.9%)
ndhJ	1 (9.1%)
rpoB	6 (54.5%)
rpoC1	4 (36.4%)
trnH-psbA	8 (72.7%)
Triplet 1 (*rpoC1+rpoB+matK*)	11 (100%)
Triplet 2 (*rpoC1+matK+trnH-psbA*)	10 (90.9%)
Triplet 3 (*rpoB+matK+trnH-psbA*)	10 (90.9%)

group species replicates together, which is a basic requirement for a barcode identification tool. From the NJ reconstruction, the three best regions presenting the highest sequence variation and the best resolution at the species level are *rpoB*, *trnH-psbA* and *matK*.

It is clear that no single region will be sufficient as an efficient and universal barcode for orchids. A multi-locus barcode, based on two or three plastid regions, seems to be the most realistic and effective solution for the identification of Mesoamerican orchids. Our results show that a "triplet" of regions would be successful with a combination of regions like *rpoB*, *matK*, *trnH-psbA* or *rpoC1*. The next step for a multi-barcode will depend on the choice of using only coding regions or including a non-coding gene like *trnH-psbA*, although this gene presents practical complications with alignment.

ACKNOWLEDGMENTS. We thank the Darwin Initiative, the Consortium for the Barcode of Life, the Alfred P. Sloan and Gordon and Betty Moore Foundations.

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Guillaume Gigot was first educated at the University of Montpellier (France) where he studied evolution and ecology. He was then awarded his diploma of engineering in agronomy and environment at "Grande Ecole" in Paris. After working on several research projects in tropical ecology and population genetics in France, he started at the Royal Botanic Gardens, Kew in 2005 as Darwin Initiative Project Officer. He is currently in charge of the coordination and management of a project regarding orchid biodiversity and DNA barcoding in collaboration with the Lankester Botanical Garden in Costa Rica.

Jonathan van Alphen-Stahl completed his BSc in Biology, Earth and Environmental Sciences at the University of Cape Town in 2001. He went on to do his honours in Botany and then completed his Masters degree in Systematics and Biodiversity Science at the University of Cape Town, with his dissertation involving the phylogenetics and phylogeography of the Helmeted Guinea fowl. He worked as biodiversity officer at the University of Pretoria before moving to the Royal Botanic Gardens, Kew. He is currently working as a data analyst on DNA Barcoding at Kew.

Diego Bogarín obtained his degree in Biology at the University of Costa Rica. He is a researcher at Lankester Botanical Garden interested in taxonomy and systematics of neotropical Orchidaceae. Recently, he is developing floristic projects for conservation in Costa Rican National Protected Areas System. He started in 2005 as Darwin Initiative Project Implementation Officer in Costa Rica for the project "Conservation and monitoring of Meso-American orchids", in collaboration with Royal Botanic Gardens, Kew.

Mark Chase received his undergraduate degree from Albion College, Michigan and his Ph.D. was from the University of Michigan (Ann Arbor) in 1985. His thesis was a monograph of *Leochilus* (Orchidaceae). He carried out post-doctoral research in molecular biology with Jeff Palmer at the University of Michigan. He then moved to the University of North Carolina and then after four year to the Royal Botanic Gardens, Kew, where he set up the program in molecular systematics. He became a member of the Royal Society in 2003 and Keeper (Director) of the Jodrell Laboratory in 2006.

Jorge Warner obtanied his master degree at the University of Costa Rica. Actually, he is the Director of Lankester Botanical Garden and currently leader of the project “Conservation and monitoring of Meso-American orchids”, sponsored by Darwin Initiative and developed in collaboration with Royal Botanic Gardens, Kew. His main interests are the biology and conservation of Costa Rican epiphytes.

Vincent Savolainen is a Plant Molecular Systematist and Deputy/Acting-Head of the Molecular Sytematics Section, at the Jodrell Laboratory, Royal Botanic Gardens, Kew. He received his PhD in Biology in 1995, from the University of Geneva, Switzerland, specializing in Molecular Phylogenetics and Evolution. His research interests include angiosperm phylogeny, Tree of Life, evolutionary processes and phylogenetics patterns, speciation and DNA Barcoding. He is currently leader of two projects funded by the Darwin Initiative for the Survival of Species in Southern Africa and Central America and several other European projects. He is presently a Fellow of the Linnean Society of London and was awarded the Bicentenary Medal of the Society in 2006.

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LAS ORQUÍDEAS DEL PARQUE NACIONAL BARRA HONDA, GUANACASTE, COSTA RICA

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Palabras Clave: áreas protegidas, Costa Rica, estudios florísticos, Orchidaceae, Parque Nacional Barra Honda

Introducción

Los trabajos florísticos constituyen fuentes de información de suma importancia pues entre otros aspectos, determinan el número de especies presentes en una región y permiten su correcta identificación (Atwood 1987, Pupulin 1998). La carencia de este tipo de trabajos dificulta futuras investigaciones en los diversos planos de la biología, la conservación y la educación ambiental. Costa Rica posee uno de los sistemas de áreas de conservación más importantes a nivel mundial. Cerca del 25% del territorio se encuentra protegido en unas 155 áreas silvestres, reservas biológicas y parques nacionales (Boza 1986). A pesar de esto, el aporte de los estudios florísticos al conocimiento de las especies de orquídeas que habitan en dichas áreas ha sido escazo y los inventarios respaldados por especímenes en herbarios son una excepción (Atwood 1987, Pupulin 1998, Gómez-Laurito & R. Ortiz 2004). La mayoría de las áreas protegidas en Costa Rica carecen de esta información, la cual se encuentra muchas veces dispersa en registros de herbarios locales o extranjeros (Jiménez & Grayum 2002, Pupulin 2003).

Materiales y métodos

El Parque Nacional Barra Honda (PNBH) se ubica en el cantón de Nicoya, provincia de Guanacaste (Boza 1986) (fig. 1). El interés de crear este parque surgió como una medida para proteger el sistema de cavernas de la región de Nicoya, originado por los eventos geológicos desde el Paleoceno Superior y Eoceno Inferior hasta el presente. Esta zona representa un área geológica muy importante para el país. (Mora 1978, Aguilar & Denyer 2001, Jaccard *et al.* 2001). No obstante, la proteción de los recursos biológicos e hídricos es también otro de los objetivos se su creación. El PNBH comprende una extensión de 2.295 hectáreas donde predomina el bosque seco semideciduo del Pacífico Norte, caracterizado por una marcada estacionalidad climática (Tosi 1969, Janzen 1983). Históricamente, estos bosques han sufrido una fuerte intervención humana producto de la deforestación y el desarrollo de las actividades agropecuarias. Por otro lado, han sido bosques poco atractivos para los orquideólogos dada su baja diversidad de orquídeas en comparación con los bosques húmedos montanos y premontanos. Ante la fragilidad de estos ecosistemas de bosque seco, únicos en el país y la carencia de datos que permitan evaluar el número de especies y su identidad, es indispensable el aporte de los estudios florísticos para llevar a cabo acciones de conservación prioritarias en esta área (Boza 1986, Janzen 1983).

El presente estudio se realiza con base en recolectas de orquídeas en los diferentes sitios del PNBH. Durante el mes de Julio del 2005 y Febrero del 2006 se obtuvieron los datos que aquí se presentan. El material recolectado fue llevado a cultivo en las colecciones vivas del Jardín Botánico Lankester, Universidad de Costa Rica. Los testigos fueron depositados en las colecciones en alcohol del Jardín Botánico Lankester (JBL) y en el Herbario Nacional de Costa Rica (CR).

Resultados

En la primera etapa del estudio se determinó que la flora de orquídeas está compuesta por 26 especies en 23 géneros. Los géneros más representados son *Epidendrun* (3 especies), *Scaphyglottis* y *Specklinia* (ambos con 2 especies). Los demás

FIGURA 1. Ubicación del Parque Nacional Barra Honda.

géneros (88.5%) están respresentados en el parque por una sola especie. El 73% de las especies son epífitas y un 27 % presenta un hábito terrestre (cuadro 1). El período de floración se concentra entre los meses de Noviembre a Abril, básicamente durante la estación seca.

Discusión

Algunas de las especies del PNBH tienen una amplia distribución a nivel nacional y habitan en toda la costa Pacífica, tanto en las zonas húmedas del sur como en las áreas secas estacionales del norte y parte del Valle Central. Estas especies son: *Epidendrum coronatum* Ruiz & Pav., *Epidendrum stamfordianum* Bateman, *Epidendrum vulgoamparoanum* Hágsater y *Scaphyglottis stellata* Lodd. ex Lindl. Tres especies, *Pleurothallis quadrifida* (Lex.) Lindl., *Scaphyglottis micrantha* (Lindl.) Ames & Correll y *Sobralia decora* Bateman presentan este tipo de distribución pero se les puede encontrar también en la parte húmeda del Caribe norte. Además, *Brassavola nodosa* (L.) Lindl., *Trigonidium egertonianum* Bateman ex Lindl., *Catasetum maculatum* Kunth, *Specklinia grobyi* (Bateman ex Lindl.) F. Barros y *Dimerandra emarginata* (G.Mey.) Hoehne, pueden distribuirse, tanto a lo largo de toda la vertiente Pacífica como en la vertiente Caribe. En el PNBH estas especies se pueden observar con relativa frecuencia en los bosques secundarios poco alterados.

Dentro de la región biogeográfica del Pacífico Norte, existe otro grupo de especies que se encuentran en las zonas estacionales húmedas y secas hacia el norte de la cuenca del río Grande de Tárcoles y alcanzan sitios estacionales cercanos al Valle Central. Este grupo presenta su límite de distribución sureño en las áreas circunvecinas al Cerro Turrubares, la cuenca del Tárcoles y el río Candelaria y su hábitat corresponde más con los bosques estacionales secos del norte (Jiménez & Grayum 2002). Dentro de este grupo encontramos *Barkeria obovata* (C. Presl) Christenson, *Cohniella cebolleta* (Jacq.) Christenson, *Cyrtopodium paniculatum* (Ruiz & Pav.) Garay, *Encyclia cordigera* (Kunth) Dressler, *Laelia rubescens* Lindl. y *Trichosalpinx blaisdellii* (S. Watson) Luer. Estos datos son apoyados por estudios florísticos en la región central y sur de la costa Pacífica, donde no se reporta la presencia de estas especies (Pupulin 1998, Weber *et. al* 2001, Jiménez & Grayum 2002). Sin embargo, esta misma área biogeográfica es el límite para algunas especies del Pacífico Central y Sur que crecen en las áreas más humedas y poco estacionales y no alcanzan una distribución más norteña. Este patrón es observado en la flora de orquídeas del PNBH y especies como *Specklinia corniculata* (Sw.) Steud., *Aspasia epidendroides* Lindl., *Ionopsis satyrioides* (Sw.) Rchb.f., *Trizeuxis falcata* Lindl., *Prosthechea abbreviata* (Schltr.) W.E.Higgins, *Campylocentrum multiflorum* Schltr. y *C. micranthum* Lindl., entre otras, se distribuyen al sur de la costa Pacífica sin alcanzar las áreas estacionales del Pacífico Norte (Pupulin 1998, Jiménez & Grayum 2002).

En el PNBH se protege la flor nacional, *Guarianthe skinneri* (Bateman) Dressler & W.E. Higgins. Su distribución es también amplia a lo largo del territorio nacional, sin embargo sus poblaciones silvestres han sido reducidas por la deforestación y excesiva recolecta. Algunas poblaciones se encuentran en el PNBH sin embargo es difícil obsevarlas aparentemente por la extracción ilegal dentro del parque.

Siete especies con hábito terrestre se encuentran en el parque. Durante el período seco, todas las plantas de estas especies presentan un comportamiento deciduo, perdiendo sus hojas durante los

CUADRO 1. Hábito y distribución general de las orquídeas del Parque Nacional Barra Honda.

Género y especie	Hábito	Amplia distribución	Influencia Pacífica	Pacífico Norte
1. *Barkeria obovata* (C. Presl) Christenson	E			x
2. *Beloglottis costaricensis* (Rchb.f.) Schltr.	T		x	
3. *Brassavola nodosa* (L.) Lindl.	E	x		
4. *Catasetum maculatum* Kunth	E	x		
5. *Cohniella cebolleta* (Jacq.) Christenson	E			x
6. *Cyrtopodium paniculatum* (Ruiz & Pav.) Garay	T			x
7. *Dimerandra emarginata* (G.Mey.) Hoehne	E	x		
8. *Encyclia cordigera* (Kunth) Dressler	E			x
9. *Epidendrum coronatum* Ruiz & Pav.	E		x	
10. *Epidendrum stamfordianum* Bateman	E		x	
11. *Epidendrum vulgoamparoanum* Hágsater	E		x	
12. *Guarianthe skinneri* (Bateman) Dressler & W.E. Higgins	E		x	
13. *Habenaria macroceratitis* Willd.	T	x		
14. *Laelia rubescens* Lindl.	E			x
15. *Malaxis aurea* Ames	T		x	
16. *Oeceoclades maculata* (Lindl.) Lindl.	T	x		
17. *Palmorchis* sp.	T			
18. *Pleurothallis quadrifida* (Lex.) Lindl.	E	x		
19. *Scaphyglottis micrantha* (Lindl.) Ames & Correll	E	x		
20. *Scaphyglottis stellata* Lodd. ex Lindl.	E		x	
21. *Sobralia decora* Bateman	E	x		
22. *Specklinia grobyi* (Bateman ex Lindl.) F. Barros	E	x		
23. *Specklinia microphylla* (A. Rich & Galeotti) Pridgeon & M.W.Chase	E		x	
24. *Sarcoglottis* sp.	T			
25. *Trichosalpinx blaisdellii* (S. Watson) Luer	E			x
26. *Trigonidium egertonianum* Bateman ex Lindl.	E	x		

E = Epífita; T = Terrestre

meses secos. Únicamente *Palmorchis* sp. y *Oeceoclades maculata* (Lindl.) Lindl. conservan sus hojas durante todo el año. Las plantas de *Cyrtopodium paniculatum* crecen en áreas rocosas con materia orgánica en bosques secundarios. *Habenaria macroceratitis* Willd. se encuentra a orillas de caminos en zonas expuestas y alteradas o en el bosque secundario. *Sarcoglottis* sp., una especie aún no identificada, crece en grandes colonias en el interior de los bosques secundarios y zonas rocosas en sitios con sombra. Las poblaciones presentan cierta variación en la coloración de las hojas y las flores. *Beloglottis costaricensis* (Rchb.f.) Schltr., una especie poco conocida y casi nunca recolectada es fácil de observar en el PNBH usualmente en poblaciones mezcladas con *Sarcoglottis* sp.

Dentro de los hallazgos más importantes está la presencia de *Malaxis aurea* Ames, una especie poco común, conocida únicamente del bosque húmedo premontano. En el PNBH se encuentra en zonas de bosque secundario en suelos rocosos con materia orgánica. Este hallazgo representa un dato ecológico nuevo para esta especie. Una especie de *Palmorchis* todavía no identificada es también un nuevo registro en términos ecológicos para el sitio y podría tratarse de una especie no descrita (Dressler, com. pers. 2005). La especie exotica y recién naturalizada, *Oeceoclades maculata* es común en ciertas áreas del PNBH y forma colonias a lo largo de áreas de bosque secundario. Su presencia se reporta para las áreas secas del Parque Nacional Santa Rosa y ha sido recolectada en el Pacífico Central y Sur (Dressler 2003).

Las acciones futuras del proyecto se concentran en evaluar otras áreas del PNBH pues es posible encontrar algunas especies que habitan en la misma región biogeográfica del bosque seco. Además, el material disponible de *Specklinia microphylla* (A. Rich & Galeotti) Pridgeon & M.W.Chase y *Trichosalpinx blaisdelli* (S. Watson) Luer, requiere de un estudio taxonómico detallado, siendo posible que estos nombres no estén correctamente aplicados al material encontrado. El presente estudio proporciona nueva información sobre los ecosistemas de bosque seco e intenta explicar la flora de orquídeas del PNBH basandose en la distribución general de las especies a nivel nacional. Esta información puede ser aprovechada por las demás áreas protegidas dentro el mismo ecosistema, las cuales comparten algunas especies que alcanzan una distribución más amplia. De esta manera se pretende iniciar un sistema que permita fortalecer el conocimiento y la protección de la flora de orquídeas de la región del bosque seco de Costa Rica.

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EPIDENDRA, THE BOTANICAL DATABASES OF JARDÍN BOTÁNICO LANKESTER AT THE UNIVERSITY OF COSTA RICA

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Introduction

It may seem somehow out of line to present a new system of botanical databases in the context of a meeting on orchid conservation, for two main reasons. Even though botanists have been rather slow in upgrading to the use of electronic databases (with some early controversy regarding the desirability of the application of electronic data processing methods to taxonomic problems as a whole, see *i.e.* Shetler 1974), the dissemination of plant information via the web has grown steadily in recent years. So, why *another* system for electronic retrieval of botanical information? On the other hand, the role of natural history collections data is perhaps better defined today as for its two-fold relevance in research and education than with respect to the practicality of information in conservation efforts. Can a system for electronic interchange of plant information be of real use as a conservation tool?

I hope that trying to answer these two questions may explain the reasons for creating Epidendra, the botanical databases system of Jardín Botánico Lankester (JBL) at the University of Costa Rica, as well as illustrate some useful characteristics of this project.

Access to the sources

For centuries, scientists have amassed information on plant life, describing and naming more than a quarter million of species on the planet. When organized in the format of floras, information included relevant data not only about morphology, but also on distribution and other aspects of plant biology. It is true that from the personal computer in his office, in any part of the world, a botanist may instantly link today to a number of powerful electronic databases, avoiding the time to correspond and to travel to botanical libraries and herbaria in order to gather the requested information, an activity that only a few decades ago would have taken months (Allen 1993). However, it may be useful to understand which kind of information is mostly available in actual databases, and how we can improve information access.

If one accesses today the TROPICOS database, launched in 1983 by the Missouri Botanical Garden (which has been a leading institution in computerizing plant information), he can find a system dealing with tens of thousand of plant names from around the world, in many cases cross-referenced with distribution maps and other non-taxonomic information. The system is designed to provide references to plant names, basionyms and synonyms, nomenclatural types, and lists of *exsiccata* for selected regions, allowing botanists to gain ready access to the authors of names, the titles of key publications and, indirectly, to the location of type specimens. This system of references has shown its relevance in floristic projects as the Flora of North America, the Floras of Panama and Mesoamerica, the Flora of Peru and the Flora of China, and it provides daily information for researchers working with tropical floras around the world, including the staff of JBL.

To restrict the field to orchids, the database Bibliorchidea, now hosted by the Swiss Orchid Foundation and operating under patronage of the University of Basel, represents the largest orchid literature database worldwide, containing most of the existing journal articles, books and preprints on orchids with over 140,000 entries. The database offers a nearly complete system of references to the titles of publications, extending the coverage not only to the original protologues but also to different types of literature quotations (for more details, see Jenny 2007). Numerous, less "institutional" databases, mainly aimed to quick orchid identification via electronic images, exist on the web, but the quality of the provided information is often not totally reliable, and they will not be considered for the purpose of this work.

One common character of the available tools for electronic retrieval of botanical information is that they provide a system of references, which supposes some facility in the direct access to the sources through libraries and herbaria services. This is often not the case in tropical countries, where facilities are insufficient, if not absent, and where the lack of historical libraries and the relatively "modern" herbaria represent a major obstacle for botanic research when concerned with the retrieval of historical information (Gómez-Pompa & Nevling 1988, Pupulin & Warner 2005).

Some steps in this direction have been made in recent years, through the digitalization of type specimens in several institutions. Noteworthy is the recently completed project of digitalization of the Oakes Ames Orchid Herbarium types at Harvard University. However, it is perhaps interesting to note that the first actions of this project were done in the framework of a cooperative effort between the Harvard University Herbaria and the University of Costa Rica, originally aimed to the digital imaging documentation of the types of Costa Rican Orchidaceae (Pupulin & Romero 2003).

One of the more crucial points to be resolved in order to achieve the goal of an open system for the retrieval of biological sources is the sociological impediment to data interchange, through the protection of copyrights and intellectual property, concerning ownership and ultimate usage of the information. Most of the valuable documents relative to the tropical flora are stored in institutions of the developed countries, sometimes jealous of the historical value of the owned sources. It is curious to note, as Conn (2003) did, that copyrights concerns are vigorously debated when the source collections are presented in a digital format, but not when available as physical collections *per se*. However, the recent agreements signed by the University of Costa Rica with the Harvard University Herbaria and with the Herbarium of the Royal Botanic Gardens, Kew, to digitally document the specimens and the associated data of the orchids from the Mesoamerican region, are an unquestionable step in the right direction.

Conservation data

Natural history collections have always contained a large amount of data providing biogeographic, ecological and biographical information through the labels affixed to the specimens, and they have been considered an indispensable resource for conservation policies, documenting what we do and do not know about the biota (Lane 1996). Nevertheless, while the threatened tropical biota is the major biological concerns of today's humankind, and the need for floristic research in the tropics is greater that in any other time in modern history, most of the global important collections are stored in developed countries. This has been an impediment to a vaster documentation of biological variation, which is required for a full understanding of living diversity, ecosystem dynamics and their conservation. Our question should be if the actual documentation of tropical biodiversity (or orchid biodiversity, to restrict to our concerned topic) is sufficient to help the conservation "movement", transforming floristic research into an actor in the conservation play. The actual figures point toward a negative answer. In a short review of the available records kept in six major herbaria relatively to 350 Costa Rican orchid species, Dressler (1996) found that 78% of the taxa were represented by less than 6 collections. Of those, 20% were based on a single collection, and for 74 species (21%) he can not find a single herbarium specimen in the herbaria sampled. The obvious incongruity is that we do not know the flora of the tropics enough to really orient conservation policies, mainly if we consider that only at most 15 percent of the life diversity on Earth has been apprehended by science, and new species are turning up constantly from the scattered expeditions to rich tropical areas.

The possibility to rapidly document the presence of some species in a given area via the access to reliable electronic data may be essential in influencing decision makers at any level, but once more the quality and efficiency of this documentation is directly associated to the amount of the available information. This quality must be increased not only by a continuous updatement of distribution records, but also providing more efficient identification and "emotive" aids, like visual databases of specimens, slides, drawings, etc., helping to match the specimen with known taxa. According to

Flecker (2000), the administration of Harvard University granted 12 million dollars to the University Library for a 5-year project aimed to build a digital library infrastructure. However, this is often not the case where funds for research and documentation are limited (as in developing countries), and the justification of scientific activity through the provision of services to the general public is probably critical.

EPIDENDRA

The past debate on biological databases has mainly focused on the best model to be used in organizing the taxonomic data from literature and other sources to avoid over-simplification and to reflect the elasticity of taxonomy as well as alternative taxonomies (see, i.e., Berendsohn 1997, Conn 2003). Even though the "unofficial" adoption of one or more of the alternative taxonomies can not be avoided in the daily work, taxonomic information may become outdated very rapidly in the tropics, and this perhaps tends to reduce taxonomic decisions in the database system to a minimum. The only alternative would be to build a system and a trained staff which avoid mistakes in the capturing and management of the information, but this would greatly increase the cost of the effort.

The main constraints to the creation and maintenance of biological databases in tropical countries have been reviewed by Gómez-Pompa and Nevling (1988) and I refer to their paper for a critical analysis. It is unfortunate to say that, with the exception of computing technology, most of these constraints have not found positive solutions. However, botanists working in tropical areas have an immense opportunity to improve our knowledge of life diversity and to provide a bridge between systematic research and the general public, incorporating to their source-based systems other data which are not accessible to their colleagues in the first world. They include field observations on species frequency and natural variation, susceptible habitats, pollination biology, relationships with other organisms, etc. But, foremost, tropical botanists have the still unachieved chance to "portray" biodiversity for the use of the public through *in studio* work, mainly based on digital imaging. Knowing something always makes it more valuable, and only what it is valued will ultimately be saved.

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