

# Darwin Initiative for the Survival of Species

Annual Report 2

Genetic diversity and management implications for high

Andean guanaco populations in Peru.

## 1. Darwin Project Information

Project Ref. Number	162/12/022
Project Title	Genetic diversity and management implications for
	high Andean guanaco populations in Peru.
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UK Contractor	Cardiff University
Partner Organisation(s)	CONOPA
Darwin Grant Value	£197, 886
Start/End dates	01/07/03-30/06/06
Reporting period (1 Apr	1 Apr 2004 to 31 Mar 2005
200x to 31 Mar 200y) and report number (1,2,3)	Report number 2
Project website	www.conopa.org
	www.cf.ac.uk/biosi/research/biodiversity/staff/dodd.ht
	<u>ml</u>
Author(s), date	Ciara S Dodd, Jane C Wheeler, Michael W Bruford.

### 2. Project Background

This is a collaborative project between Cardiff University, UK and CONOPA (Coordinadora de Investigación y Desarrollo de Camélidos Sudamericanos), a Peruvian NGO based in Lima, Peru. Specifically, the project aims to address issues relating to the conservation management of the Peruvian guanaco population through direct surveys and genetic analysis. By characterising the demography and genetics of these fragmented populations we will enable a rational management programme to be established. This will result from a Population and Habitat Viability Assessment (PHVA), for the Peruvian guanaco through a workshop of all stakeholders, to be held in Lima at the end of the project.

# 3. **Project Purpose and Outputs**

The main purpose of the project is to characterise the demography and genetics of highly endangered and fragmented Peruvian guanaco populations to enable the establishment of a realistic conservation management plan in the final months of the project. In order to achieve this goal, non-invasively obtained faecal samples from six Peruvian guanaco populations were collected by the CONOPA team during this reporting period, and analysis of the samples was begun in the laboratories in Lima and Cardiff.

The second purpose of this project is to build capacity in conservation genetics in Peru. This is being achieved by providing intensive training for two Peruvian scientists, both qualified veterinarians working in CONOPA, in modern molecular genetic techniques, analysis and conservation biology. One trainee, Jorge Rodriguez (JR) completed his second six-month training period in Cardiff on 31<sup>st</sup> March 2005. The second trainee, Katheryn Yaya (KY) will begin a six-month training period in Cardiff in July 2005. In addition, both JR and KY have received training in the laboratory in Lima during the last year and in addition, JR completed the final year conservation biology module, run by Mike Bruford (MB) in Cardiff in 2003.

The third goal of the project is to train a cohort of Peruvian scientists in conservation biology and population and habitat viability analysis. This training will be in the form of two, one month-long courses, the fist of which was run from 14<sup>th</sup> June to 9<sup>th</sup> July 2004 in Lima. The second course will begin in June 2005, in Lima. Thirteen students were enrolled on the course, as well as JR and KY, all of whom obtained a diploma upon completion of the course assessments.

Alterations were made to the original project timetable at the beginning of the project and were discussed in detail in the first annual report. The effect that these have had on the operational plan for year 2 is as follows: The UK post-doc Ciara Dodd (CD) and JR travelled to Peru in March 2004 rather than January and CD returned to the UK in September 2004 rather than in July as originally scheduled. JR delayed his return to Cardiff until early October 2004 to accommodate a field expedition in September to Arequipa, so that he could bring the maximum number of guanaco samples with him to Cardiff for analysis. JR travelled back to Lima on 1<sup>st</sup> April 2005. The conservation biology course was held in Lima, beginning on 14<sup>th</sup> June, two weeks later than initially scheduled. Otherwise, the project is on schedule.

#### 4. Progress

This Darwin Initiative project began on 1<sup>st</sup> July 2003 and up to the beginning of this reporting period the project has progressed as follows. The project launch in the host country, Peru, consisted of a high profile series of meetings and presentations involving the project team, hosted by the Peruvian Congress, the British Embassy, Lima and the School of Veterinary Medicine, San Marcos University. Subsequent progress up to the beginning of this reporting period has been in accordance with the revised baseline timetable outlined in the six-month report. In brief, JR completed his first six month training period in Cardiff and attended a 10 week undergraduate conservation biology course run by MWB at Cardiff University. In Peru, KY developed a sampling strategy to incorporate observational information on guanaco behaviour and faecal sample collection. In the Lima laboratory, KY worked on standardising DNA extraction and amplification protocols for faecal samples. Due to significant difficulties encountered in obtaining permits from INRENA to allow full scale sampling to commence, fieldwork was delayed until April 2004.

The progression and achievements of the project within this reporting period, 1<sup>st</sup> April 2004 to 31<sup>st</sup> March 2005 are discussed below.

#### Field work

The fieldwork element of this project involves collecting faecal samples from guanaco living in six isolated populations (Map 1) which encompass the geographic range of the guanaco in Peru.

The revised schedule for sample collection was set out in the first annual report (summarised in table 4 below). However, continuing difficulties in obtaining a permit from INRENA to allow sample collection to commence resulted in the first field expedition being delayed until mid-April 2004. However, subsequent sampling has been successful and was ahead of schedule in September 2004.

A preliminary expedition was carried out to obtain samples at a guanaco chaccu in Huallhua in 2003. However, it has always been the intention of this project to use non-invasively obtained material such as faeces for genetic analysis, since adequate techniques for the capture of guanaco have not been developed, but more importantly for a conservation centred project, this minimises stress to the animal whilst maximising the information that can be obtained using modern molecular methods to extract DNA from faecal matter. Using a non-invasive approach is not without difficulties, both in terms of sample collection and genetic analysis, which the field and laboratory teams have had to overcome.

COR C 4 NOR DRIES SAN NARTIN ; BRASIL Calipuy (La Libertad) 2 HÁRIN CALLAD Huallhua (Ayacucho) Chavin (Ica) Machahuay (Arequipa) TEST MAR La Capilla (Moquegua) Vila Vilani (Tacna) ALTUNA IN NITHOS CHILE THE REPORT OF A DESCRIPTION OF A DESCRIP Guanaco Distribution in Peru (1996 Census) Populations sampled Guanaco populations Marcn 2005).

Map 1: Distribution of the guanaco in Peru based on the 1996 census (CONACS), and populations sampled for this project.

Proposed sampling Date	Actual date of sampling	Location and number of samples collected
April and May 2004	11 <sup>th</sup> -18 <sup>th</sup> April	30 samples collected from Huallhua, Ayacucho, in addition to 12 blood samples collected in a chaccu in November 2002.
Mid July to end August 2004	16 <sup>th</sup> May-10 <sup>th</sup> June	34 samples collected from Reserve of Calipuy, La Libertad
Sept 2004	27 <sup>th</sup> July-06 <sup>th</sup> August	23 samples collected from Chavin, Ica
October2004	22 <sup>nd</sup> August- 01 <sup>st</sup> September	38 samples collected from Machahuay, Salamanca, Yarabamba and Reserve of Salinas, Arequipa.
November 2004	14 <sup>th</sup> -17 <sup>th</sup> December	07 samples collected from La Capilla, Moquegua
December 2004	03 <sup>th</sup> -12 <sup>th</sup> December	10 samples collected from Vila Vilani and Candarave,Tacna

Guanaco primarily live on the western slope of the Andes where they are found at elevations from sea level to above 4,000 meters in the high Andean puna. They are spread across areas of remote, often precipitous and inhospitable terrain where there is little or no human presence. Since their numbers are relatively small, a population may cover a vast territory. In order to obtain faecal samples, which need to be fresh and from different animals to allow population genetic analysis, the number of people required to collect them proved to be larger than originally anticipated. Typically, four to six people were involved in each expedition working in teams of two or three and maintaining contact by walkie-talkie. A spotting scope and binoculars were used to observe guanacos and a record was made of the animal's behaviour, family grouping and position using a GPS data logger. When an animal was observed to defecate, one member of the team went to collect the sample, being directed to the sample by the other member of the team who was observing through the binoculars and guided the sample collector to the faecal deposit using the walkie-talkie. In this way, every attempt was made to relate the sample to the animal from which it came and that the sample was fresh. However, in practical terms, this method of sample collection is time consuming. It can take days to find and observe the animals, and getting to the point where the sample has been deposited can be very difficult and time consuming since observations were often made over relatively long distances in difficult terrain. This potentially has an implication for sample quality since the strong UV at high altitude could rapidly degrade the DNA on the surface of the faeces, although over the timescales involved in this study it is unlikely to be problematic. Every attempt was made to reach the sample quickly and to only collect faeces that still appeared moist and fresh. On collection, the faecal pellets are preserved in 100% ethanol in marked plastic tubes. Given the impossibility of obtaining samples from live animals, the procedure described above is the best solution available for obtaining DNA samples.

Due to the scarcity and remoteness of guanaco populations, and frequent lack of reliable information concerning their location and facilities available in the area, it was often difficult to predict the duration of expeditions, and adequately prepare supplies and provisions for members of the field team, who had to take or carry sufficient provisions and kit with them. As sampling progressed, the team learned how best to prepare and efficiency increased with experience, something that could only be learned in the field.

In addition to collecting faecal material, the field team conducted interviews with members of the local communities using a standard questionnaire that was developed for this purpose. Information was gained about the guanaco population in the community area. Information about the project and the remit of CONOPA was also disseminated within the communities where sampling took place.

Besides sampling guanaco populations, CONOPA and CONACS organised a vicuña chaccu in Catac (Ancash) on 3<sup>rd</sup> September 2004. This was attended by the CONOPA team as well as CD and Stephen Casey (SC) a colleague form Cardiff who volunteered on the project between July and September 2004. Before the chaccu, CONACS and Jane Wheeler (JCW) and Domingo Hoces (DH) from CONOPA gave presentations about the work of CONOPA with camelids, the utility of genetics in vicuña population biology and management and in particular, the aims and objectives of the guanaco DI project were discussed. The vicuña samples collected in the chaccu were analysed in Cardiff with mtDNA and microsatellite markers, and the results have been presented to CONACS. They will also be presented to the scientific community and authorities in Huaraz (Ancash) and the community of Catac on 12 May 2005. The Ancash vicuña population is located at the northern extreme of vicuña distribution and had never previously been studied. The results indicate a close relationship between Ancash and populations located in Junin, pointing towards the existence of a single genetically and geographically distinct vicuña population distributed along the Cordillera Blanca, Huayhuash and Huaytapallana mountain chain, located to the north and west of a second distinct population. The results have provided a clearer definition of Peru's northern vicuña populations and their boundaries.

#### Laboratory work

Guanaco blood samples were collected from Huallhua in 2002 and from guanacos in Lima zoo in 2003. These twelve blood samples provide good baseline data for Peruvian guanaco and served as good controls for testing and optimising laboratory methods and to act as positive controls for PCR with faecal extracts. The laboratory in Lima already has established blood extraction protocols that are used in their other areas of work. However, since the majority of guanaco samples being used in this project are of faecal origin, stool extraction protocols needed to be established and refined.

Although faecal extraction protocols had been used in the CONOPA laboratory in the past, they had only proved successful for the amplification of mitochondrial DNA. Microsatellites are more difficult to amplify from faecal material since they are found in nuclear DNA, which is present in only two copies per cell compared to hundreds per cell for mitochondrial DNA, therefore faecal extraction procedures needed to be modified for microsatellite analysis. KY worked on modifying the stool extraction protocol, specifically by only using the outer surface of the faecal pellet for extraction, increasing the length of some protocol steps and reducing reagent volumes to increase DNA concentration in the final extract, whilst maintaining extract quality. Following these modifications, it is now possible to amplify microsatellites and mtDNA from faecal DNA in the laboratory in Lima using this standardised protocol. However, DNA extracted from the guanaco samples collected in Calipuy consistently failed to amplify in PCR reactions both for mtDNA and microsatellites in Lima and in Cardiff. In Cardiff, the extraction protocol was modified further, increasing the initial incubation period and decreasing the final elution volume which resulted in successful amplification of mtDNA and microsatellites for the samples tested.

Building on work that CD and JR began in Cardiff, to multiplex microsatellites on silver stained gels, JR and KY have been working to optimise a system of multiplexed microsatellites in the lab in Lima under the supervision of CD. Following the successful amplification of multiplexes visualised on silver-stained gels, it became clear that some of the multiplexes needed redesigning to allow for differences in the techniques of visualisation of the loci between the fluorescent ABI methodology used in Cardiff and the silver-stain methodology used in Lima. The

structuring of multiplexes for silver-stained gels has to be revised due to the nature of the technique which means that co-amplified loci need to be within a smaller size range than for the fluorescent approach so that consistent separation of bands of different sizes can be achieved. In addition, loci that are co-amplified must not have overlapping allele size ranges otherwise accurate scoring of the loci will not be possible. In Cardiff we have tested many different combinations of loci and are confident that within the loci we are using, triplexes (three primer pairs amplified in the same PCR reaction) of many primer combinations will be successful with silver stained gels. JR and KY are continuing with this work in the laboratory in Lima.

In Cardiff we have had difficulties amplifying long fragments of the mitochondrial control region (d-loop) of approximately 650 bp in length from faecal DNA extracts. This problem was overcome for some samples, but the resultant sequences were not of good quality therefore two shorter overlapping fragments were amplified both of which work well and produce good quality sequences. Up to the end of this reporting period we have sequenced d-loop fragments for four populations of Peruvian guanaco. We are currently working on developing a DGGE or SSCP methodology for separating sequence haplotypes. Samples will be run to test this method for which we already have sequences and therefore know the sequence polymorphisms. It is expected that this work will continue into the third year of the project. This technique will allow the identification of guanaco mitochondrial haplotypes, visualised on gels stained with silver without the need for sequencing, the facility for which does not currently exist within the laboratory in Lima.

During the last six months, JR has received extensive training in mtDNA PCR amplification and sequencing as well as sequence editing and analysis. Throughout the process, the techniques have been demonstrated to him and until reaching proficiency, he has conducted these activities under the supervision of CD. CD has provided advice and guidance and troubleshooting throughout the process. JR has received preliminary training in genetic data analysis, but more in depth tuition will be given once the sequence data for the guanaco is complete. Both JR and KY will be trained together in the next reporting period and contact will be maintained by email.

As a result of the training received by JR in UK, he is using his knowledge to teach scientists within the laboratory in Lima and to deliver lectures on molecular markers at San Marcos University, Lima. Building on JR's work on parentage testing of alpacas, which he conducted in Cardiff last year for his thesis, a new collaborative project, funded by IAEA, between Cayetano, CONOPA and IPEN has been established to initiate parentage testing for the government run alpaca registry.

JR has also applied the techniques that he has learned to work on two new projects in the lab. Firstly, he has developed primers that enable the sexing of camelid samples which have come from individuals for which the sex is unknown. To do this, JR amplified sex chromosome fragments using general bovid primers, sequenced the fragments and then identified sex specific sequence polymorphisms. Specific PCR primers were then designed to amplify these fragments, showing one band for female and two bands for male visualised on agarose gels. These primers work in guanaco, vicuña, alpaca and llama and will be of great use for the work of CONOPA.

Secondly, JR has been using RAPD markers to identify bands that are correlated with fibre quality in genetically 'pure' camelids. The results of this work are intended for form the basis of a grant application by JR and CONOPA to further investigate fibre quality in camelids. Again this work will be tremendously beneficial for the wok of CONOPA now and in the future. These activities demonstrate that JR is acquiring the research skills necessary for him to develop and establish active research and training programmes and to be fully independent as a scientist at the end of the project. This fulfils one of the key objectives within the logical framework of this project.

KY has obtained funding from San Marcos University for research on Molecular Etiopathogenesis of Enterotoxemia in Alpacas. In this project she will be applying

skills learned under the Darwin Project to help resolve a major health problem in Peru's alpaca population.

All progress within the project has been monitored in Cardiff and in Lima through weekly lab meetings held by MWB and JCW respectively. These meetings provide the opportunity to outline progress and discuss problems and future work with the project team and in Cardiff provides the opportunity to discuss ideas with other lab members and tap into their expertise where required. Contact was maintained between CD (whilst in Peru) and MWB via a weekly email update outlining the progress and or issues arising during the week. In addition there have been a number of specific project and planning meetings with MWB and JCW.

#### Conservation Biology Course

Planning of the course began with the arrival of CD in Lima on 22<sup>nd</sup> March 2004 and involved the whole project team. The course was held between 14<sup>th</sup> June to 9<sup>th</sup> July 2004 and hosted by Universidad Cayetano Heredia in Miraflores, Lima. The structure of this course was extended from the framework of previous successful courses run as part of DI projects in Gabon (08/044) and Malaysia (09/016). Whilst overall course structure and core lecture topics were maintained for this project, specific examples and activities were tailored to the issues relevant to conservation within the different habitat zones of Peru.

The core course material was delivered by CD and MWB in an intensive first two weeks, consisting of lectures, practical exercises and discussion. The lectures covered topics ranging from an introduction to conservation biology, threats to biodiversity, small population biology and extinction, to the application of genetics to conservation and population and habitat viability assessment. Students were assigned the task of finding as much information as possible about an endangered species of their choice which they would then use in a practical exercise on PHVA using the programme VORTEX at the end of the second week. All the core lectures were delivered by CD and MWB in English, but each student was provided with a Spanish translation of each presentation. The Spanish translation of the course will be provided in the Final Report.

The second two weeks of the course consisted of talks by invited speakers covering all aspects of conservation projects in practice in Peru. Speakers were invited from governmental, university and conservation organisations and a variety of NGOs within Peru. Three special topic days were also organised which were open to members of the public. The topics of these were: 1. Legal Framework and Politics of Conservation; 2. Conservation in Practice in Peru; 3. Vicuña and guanaco Conservation. Notable speakers who gave presentations at these sessions were invited from CONAM, IUCN, INRENA, Peruvian Congress, Conservation International and WWF amongst others. These three days were well attended and received by those present. These open days were advertised in the local newspapers, at the Universidad Cayetano Heredia, Faculdad de Medicina Veterinaria, Universidad de San Marcos and on the CONOPA website. Posters and leaflets were produced for advertising the course, all of which featured the Darwin and project logos.

A daylong visit to the International Potato Centre (CIP) was made in the third week of the course where the students learned about gene banks, conservation and sustainable utilisation. This provided the opportunity for the students to visit hightech and high throughput laboratories and to observe the application of genetics to conservation from an agricultural perspective, providing a good contrast to the emphasis of the rest of the course.

A three day field course was organised to Reserve Nacional de Paracas in collaboration with INRENA. During the field course, the students were given lectures by INRENA staff working in the national park, had the opportunity for discussions with them about conservation issues in the park and were given guided educational tours around the park. In particular, the difficulties of integrating effective conservation management in the reserve with the needs of local people using and/or

living within the reserve were highlighted. In addition, a special presentation was given to the students by the members of the CAMISEA project in Pisco. This team is responsible for monitoring the impact of a large cross country gas pipeline which begins at a location situated just outside of the Paracas Reserve. This visit prompted intense discussion between the students and the CAMISEA project representatives.

Applicants for the course were invited from Peruvian NGOs, international conservation organisations and government departments such as INRENA and CONAM. In total, 13 students were enrolled onto the course, the breakdown of which is detailed in the 18 month report, after their applications had been evaluated by JCW, DH and CD. In addition JR and KY enrolled on the course and all 15 students were awarded diplomas. This required a minimum of 70% attendance, active participation in course activities and discussion which was monitored by CD, as well as satisfactory completion of three assessment exercises; i) individual presentation about an endangered species, ii) group presentation about a protected area, and iii) an individual presentation of a scientific paper. Through the discussion and assessment process delivered through this course the students were able to demonstrate increased learning and show that they had digested and understood the messages and information delivered to them throughout the course. This meets one of the key objectives detailed in the logical framework.

An important outcome of the course has been the establishment of a web by the students who participated through which they keep in touch and are working together in many different areas of conservation. Additionally, as a result of the PVA work, at least three interdisciplinary research proposals have been prepared and submitted to different funding agencies for work on conservation of the spectacled bear, the torrent duck and peccary.

#### Other Progress

Additional general points that are worthy of note are that within CONOPA, the other laboratory members, have benefited from the training given to JR and KY who have been transferring skill to them. There has also been an increase in the willingness and ability of them to understand English and several members of the lab enrolled in English classes in 2004. This has meant that they are more able to access scientific literature in English language journals which is increasing the scope of their scientific knowledge base. In addition several members have been keen to develop projects with and forge links with the student on the course.

Capacity has been built in CONOPA through the acquisition of a number of conservation biology and genetics text books, which were purchased by CONOPA or privately by JR, for use in CONOPA, during his visit to the UK.

The projected operational for the next reporting period is outlined in table 5 below.

Date	Activity
April and May 2005	CD to complete mitochondrial DNA analysis of remaining populations and write first paper
Early June and	CD to return to Lima to finish course preparations.
July 2005	KY to return to Cardiff with CD to begin
Mid June to Mid July 2005	MWB arrives in Lima. Second Conservation Biology Course
mid July 2005 to mid January 2006	KY travels to Cardiff for 6 months intensive training in population genetic analysis in the laboratory.
October 2005	KY takes MWBs final year conservation biology course and is assessed.

### Table 5: Operational Timetable for Year 3 (1<sup>st</sup> April 2005 to 31<sup>st</sup> march 2006)

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

Issues raised in the review of the first annual report have been addressed here and in the 18 month report. In particular, discrepancies in the budget and processes of monitoring and evaluation were discussed in 18month report.

In this report we had attempted to provide a more descriptive analysis of impacts of the project in the host country and have provided a more detailed discussion of the exit strategy. Maps and figures have also been incorporated into the report as requested by the reviewer.

#### 6. Partnerships

Continuing the collaborative link with Juan Carlos Marin (JCM) from Universidad de Chile, JCMs vicuña samples have been genotyped to build into the Peruvian dataset from the previous Darwin vicuña project (N251). These results will be submitted to a peer reviewed journal shortly.

Collaboration with JCM has also enabled the addition of important populations of guanaco that would otherwise have been impossible to sample, for example populations from Bolivia. Additional collaboration with Ricado Baldi of CENPAT, Argentina, to incorporate guanaco samples from Patagonia as well as data generated on guanaco in the DI vicuña project (N251) will further enhance the outputs of this project.

CONOPA has signed a formal agreement for collaborative research with UPCH, Universidad Peruana Cayetano Herredia. This agreement is, in part, a direct outcome of UPCH's hosting the Conservation Biology course, although there are long standing ties between both organizations. In April 2005, CONOPA, UPCH and the Peruvian Atomic Energy Agency, IPEN, initiated a joint project financed by the International Atomic Energy Agency, IAEA. The purpose of the project is to establish paternity testing using microsatellite DNA for use in Peru's alpaca registry. The project will provide CONOPA with much needed new laboratory equipment, expendable supplies and training for staff. Members of CONOPA are also collaborating in research on the guinea pig genome with Dr. Oswaldo Ramirez at UPCH, with the goal of determining and preserving biodiversity of the different domestic varieties extant in Peru. Another project with Dr. Jose Espinosa of UPCH, to create a cDNA library for alpaca skin, has reached the second funding round at the International Centre for Genetic Engineering and Biotechnology, ICGEB, Trieste.

CONOPA is in the progress of rewriting its statutes in order to expand its research area to include conservation genetics in all species, not just in South American camelids as it now stands. Research projects on the spectacled bear, peccary and tapir are in preparation.

#### 7. Impact and Sustainability

During this reporting period a number of activities have promoted the project within the host country and elsewhere.

During the course, a special topic day open to the public was organised on vicuña and guanaco conservation in Peru. At this seminar, presentations were given on the conservation biology, genetics, the outcome of our previous Darwin project on vicuña and the current guanaco project, and the role of these projects in insuring the conservation and survival of the species in Peru. In September JCW spoke about the Darwin Project and CONOPA's research at the SVII Congreso Nacional de Ciencias Veterinarias in Tacna, Peru and DH attended an international meeting on Sustainable Utilization of the Guanaco in Bariloche, Argentina. Between 21<sup>st</sup> and 24<sup>th</sup> September CONOPA and the III Peru Foundation organized a course on camelid genetics which was held at Peru's National Science Foundation, CONCYTEC, and attended by university professors from all over Peru. Michael Bruford attended as special guest speaker. The work of the Darwin Guanaco 1 project and the previous Darwin Vicuña project were discussed in deatail.

On 5<sup>th</sup> October, the Darwin Guanaco 1 project and the previous Darwin Vicuña project formed part of a special presentation made by JCW at a forum on research in molecular genetics organized by the Peruvian Atomic Energy Agency, IPEN, and the International Atomic Energy Agency, IAEA., in Lima. Between 25<sup>th</sup> and 31<sup>st</sup> October JCW observed guanacos and consulted with colleagues concerning their conservation and management at Torres del Paine National Park in Chile, and stopped in Santiago on her return to work with JCM during the first days of November.

On 6<sup>th</sup> November JCW gave a lecture on Molecular Genetics and Conservation of the South American Camelids, including a full description of the work done on both guanaco and vicuña under both Darwin projects, to Peru's largest scientific organization at the Colegio de Ingenieros in Lima. The talk was well attended, bringing together both specialists and interested parties from the government, universities and private organizations. On 9<sup>th</sup> November JCW gave a talk about the Darwin research projects and vicuña and guanaco conservation at Peru's British School, Newton College. On 26<sup>th</sup> November, JCW met with Peru's Minister of Foreign Affairs to help in preparations for the XXI Reunion Ordinaria del Convenio de la Vicuña, the Andean Vicuña Treaty, which was held in Lima between 29<sup>th</sup> November and 3<sup>rd</sup> December.

Information on the project and the broader work of CONOPA has been disseminated to all the communities where sampling is conducted. Members of the project team will return to these communities to present the findings the end of the project and to discuss the results with the local community members.

JR travelled to the 4<sup>th</sup> International Symposium on South American Camelids meeting in Goettingen in Germany in October 2004 to present a poster on paternity testing in alpacas. In addition, he also gave an oral presentation on vicuña genetics and the DI guanaco project for CD who was unable to attend due to ill health.

CONOPA has been consulted directly about implementing the results of their genetic research into law, by the director of National Parks in Peru, by the head of Consejo Nacional de Medio Ambiente, CONAM, by the Technical Director of Consejo Nacional de Camelidos Sudamericanos, CONACS and various Congressmen. The desire to incorporate the outcome of our research from the previous Darwin Vicuña project into law, as well as to set up the framework for protecting the guanaco based on our research is a direct measure of the impact which we have had.

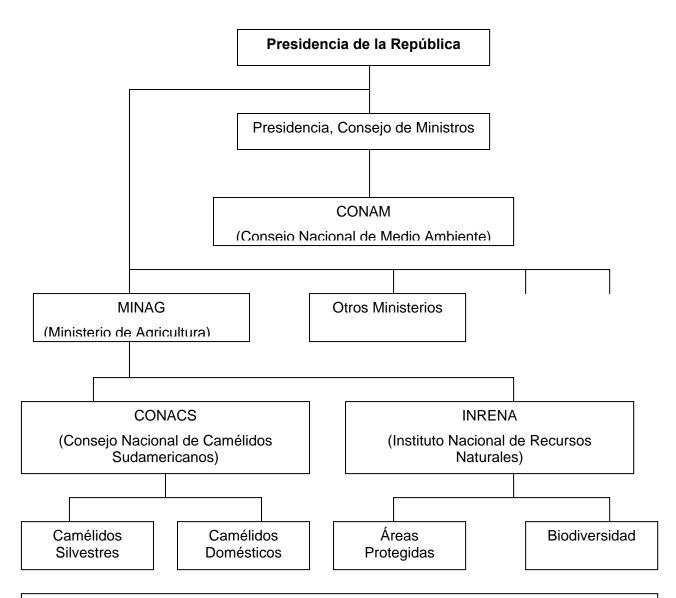
CONOPA continues to be called as a consultant by government authorities concerning conservation of the vicuña and guanaco. There is constant communication between CONOPA and CONAM (National Environment Council), the Peruvian authority concerning compliance with the CBD. At the request of CONAM, INRENA and Conservation International, JCW met with the Peruvian Minister of Foreign Affairs prior to the Andean Vicuña Treaty meeting in Lima in November to explain to him the results of our Darwin funded research on both the vicuña and the guanaco and the importance of those results in planning for conservation of both species. Under Peruvian law the guanaco is considered endangered, but it is continually lumped together with the vicuña concerning management plans, something which is totally inappropriate. The current administration at CONACS views the vicuña (and the guanaco) as an animal which should be bred in captivity and is attempting to establish a national plan to justify this goal. Captive breeding is specifically prohibited by the Andean Vicuña Treaty and also goes against the

conditions put in place by the US Fish and Wildlife Service when they accepted the CITES classification of the vicuña in Appendix II and opened the US market to the sale of legally shorn Peruvian vicuña fibre in 2002 (Federal Register Vol. 67, No. 104. 30<sup>th</sup> May 2002, 50 CFR Part 17:37695-37723). In making their decision, the USFWS paid special attention to the results of our first Darwin project on vicuña and stated that among other conditions, they expected Peru to manage their vicuña populations according to the results of our research, also a requirement of the Andean Vicuña Treaty, and something which has not been done. Clearly CONACS present position emphasizing rearing vicuña in captivity goes directly against this expectation and JCW's meeting with the Minister of Foreign Affairs just prior to the Andean Vicuña Treaty meeting dealt directly with this problem. After CONACS subsequent presentation of their new vicuña and guanaco management plan at the treaty meeting, protests were filed by various governments, and the project has apparently been put on hold. Getting to this point without offending the CONACS authorities became virtually a full time undertaking for JCW and DH starting in September 2004.

As a result of these problems, and specifically given the ignorance of the authorities at CONACS concerning the situation of the guanaco in Peru, CONAM and CONOPA are presently planning a national campaign to highlight the plight of the guanaco and educate both governmental organizations and the public. The importance of CONAM is that it is directly responsible to the president of the council of ministers and the president of Peru. In the area of South American Camelids, their role is to insure that the activities carried out by CONACS (National Council for South American Camelids) and INRENA (National Natural Resource Institute) fulfil Peru's legal obligations. At a lower level, within the Ministry of Agriculture, CONACS and INRENA each run two programmes which involve the guanaco and vicuña, wild and domestic animals at the former; protected areas and biodiversity at the latter. Unfortunately the legislation is not clear concerning the remits of CONACS and INRENA and this creates problems. Up until two months ago INRENA was responsible for issuing CITES permits, but this has now been passed to CONACS creating problems and perhaps interminable delays in the permitting process. This could have a potential impact on our ability to send samples to the UK for study on schedule. Given this situation CONAM must be looked to negotiate between the conflicting interests and place some order in the matter. As things stand, CONAM will play a crucial role in completion of the goals of the Darwin project and drawing up of the management plan.

As requested by the reviewer, a schematic of the relationships between CONOPA and the various government agencies responsible for conservation, environment and camelids is given below (Figure 1).

Figure 1: Schematic representation of the interrelationships between Peruvian government departments responsible for conservation and camelids and CONOPA.



CONOPA – Coordinadora de Investigacion y Desarrollo de Camelidos Sudamericanos

CONOPA is a NGO conducting research in the area of South American Camelids. CONOPA works on the basis of formal agreements (convenios) with CONACS and INRENA and under the patronage of CONAM, the institution responsible for overseeing compliance with international conventions (CBD etc.) signed by Peru.

#### Exit strategy

The major scientific and conservation aim of this project is a Population and Habitat Viability Assessment (PHVA) is planned for April 2006, three months before the end of the project. The purpose of the PHVA is to use the population demographic and genetic data obtained throughout the project to formulate a management plan for the Peruvian high Andean populations of the guanaco. This PHVA will be conducted as a workshop to which the key stakeholders will be invited and all stakeholders will be involved in the production and implementation of the management plan. However in addition to this it is envisaged that by the end of the project, CONOPA will have the

facility to produce high quality genetic data using the techniques the two trainees learned during the course of the project and have an increased capacity to develop and complete new projects.

In order to ensure the ongoing capacity within CONOPA for addressing conservation and scientific issues of camelid conservation in Peru, it is specifically anticipated that CONOPA will be able to achieve the following functions by the end of the project.

- a. High quality reproducible genetic data can be produced for population, phylogeographic and phylogenetic analysis from good quality and challenging DNA samples. They will be able to produce this data using the techniques available to them in their laboratory such as silver staining, but will also be able to use more modern methods if that facility becomes available in CONOPA in the future and will be able to train others in these techniques. JR and KY will be able to analyse this data using appropriate statistical methods. They will have the ability to apply their knowledge to using new types of analysis and will know where to find information and programmes to enable them to do this.
- b. JR and KY will be able to work independently and train others in laboratory methods. JR already has final year project students in Lima to whom appropriate training needs to be given. JR also teaches some classes in molecular biology in the university. In Lima KY has provided laboratory training and supervision of thesis students who have been working on projects such as sexing the *Pava aliblanca*, an endangered.
- c. JR and KY will be able to use their genetics expertise to solve problems and apply this knowledge to new problems and write grant applications. JR and KY are writing grant applications to the International Science Foundation obtain funding for further research within CONOPA. KY has already obtained funding for a research project on Molecular Etiopathogenesis of Enterotoxemia in Alpacas.

#### 8. **Post-Project Follow up Activities** (max 300 words)

Not applicable.

#### 9. Outputs, Outcomes and Dissemination

#### Outputs

All the agreed outputs for the period have been achieved as well as some additional ones as follows.

(3) Fifteen Peruvian students were enrolled on the course (including KY and JR) rather than the anticipated number of ten. Although not included as an output, a number of CONOPA staff and associates attended many of the course lectures and activities, but did not complete the assessment or receive diplomas.

(6a) Both JR and KY received training in the laboratory in Lima, as did a number of other CONOPA staff.

(8) The UK post-doc (CD) spent an extra five weeks in Lima between 23<sup>rd</sup> March 2004-10<sup>th</sup> September 2004. In addition to this a British ex-post-doc from MWBs lab in Cardiff (SC) came to Lima to volunteer on the project for 10 weeks between 3<sup>rd</sup> July 2004-10<sup>th</sup> September 2004.

(15a) A press release was issued to advertise the public sessions in local newspapers.

(20) Various text books were purchased by CONOPA or privately by JR for use by CONOPA and for use by course participants.

#### Outcomes

An important outcome of the course has been the establishment of a web by the students, speakers and professors through which they keep in touch and have begun working together in many different areas of conservation. At least three interdisciplinary research proposals have been prepared and submitted to different funding agencies for work on conservation of the spectacled bear, the torrent duck and peccary.

#### Dissemination and target audiences

Many of these issues are discussed in other sections especially section 9. However in general, it is envisaged that the project goals and findings will be disseminated to as broad an audience as possible. The course specifically targeted young professionals working in conservation biology, but the public lectures were freely available to anyone who wished to attend. Information is also disseminated to the local communities on whose land the guanaco live and where sampling takes place, since without their support, the goals of the project will not be fulfilled.

Code No.	Quantity	Description	
3	13 +2	Peruvian scientists receiver diplomas from conservation biology course (+2 = JR and KY)	
5	2	JR was trained in the labs in Lima and Cardiff, and KY was trained in the lab in Lima for 6 months by CD and for six months by JR	
6b	2x5months	JR and KY received training in laboratory techniques in	
	1x6months	Lima	
		JR receives training in Cardiff for 6months	
7	1	A CD of training materials from the course (core lectures and guest lectures and references and other useful information) was disseminated to all course participants and course speakers.	
8	25 wk	CD providing training in laboratory to CONOPA staff (march-Sept) and preparation for course	
	2wk	MWB to Lima for course	
	10 wk	SPC, a molecular ecologist, voluntary work on the project in the lab, course and fieldwork.	
15a	1	Press release for course public open days in Peruvian national newspapers.	
20	6	Text books purchased on conservation biology and genetics for use by CONOPA and student s on the course.	

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

#### Table 2: Publications

Type *	Detail	Publishers	Available from	Cost £
(e.g. journals, manual, CDs)	(title, author, year)	(name, city)	(e.g. contact address, website)	

#### **10. Project Expenditure**

• Please expand and complete Table 3.

# Table 3: Project expenditure during the reporting period (Defra Financial Year01 April to 31 March)

Item	Budget (please indicate which document you refer to if other than your	Expenditure	Balance
	project schedule)		

 Highlight any recently agreed changes to the budget and explain any variation in expenditure where this is +/- 10% of the budget.

There is an apparent under spend on travel in this financial period. This is due to the alteration of the original project timetable which has meant that some flights have occurred later than planned and cost more than initially anticipated. This also resulted in an over spend on travel in the previous period, which on balance cancels out the under spend observed here.

#### 11. Monitoring, Evaluation and Lessons

Monitoring within the project occurs on a regular and ongoing basis both in Lima and in Cardiff. Regular email contact is maintained between members of the project in both locations and provides the opportunity to rapidly address any issues that may need to be discussed or require action. Regular weekly lab meetings occur in Cardiff and Lima with the UK and Peruvian project leaders. In Cardiff a regular journal club and seminar series takes place and provide an environment in which any issues can be discussed with the project leader and also with other members of the lab, drawing on their areas of expertise. Day to day monitoring of the trainees performance in the laboratory is conducted by CD. Typically, the trainee is assigned a particular task and once completed CD discusses the results and any problems that may need to be addressed, although troubleshooting advice is provided throughout the process as required. In this way the trainee gains the maximum amount of experience and proficiency for the techniques which he/she is learning.

# 12. OPTIONAL: Outstanding achievements of your project during the reporting period (300-400 words maximum)

#### I agree for ECTF and the Darwin Secretariat to publish the content of this section

In this section you have the chance to let us know about outstanding achievements of your project over the year that you consider worth highlighting to ECTF and the Darwin Secretariat. This could relate to achievements already mentioned in this report, on which you would like to expand further, or achievements that were in addition to the ones planned and deserve particular attention e.g. in terms of best practice. The idea is to use this section for various promotion and dissemination purposes, including e.g. publication in the Defra Annual Report, Darwin promotion material, or on the Darwin website. As we will not be able to ask projects on an individual basis for their consent to publish the content of this section, please note the above agreement clause.

Annex 1 Report of progress and achievements against Logical Framework for Financial Year: 2004/2005

Project summary	Measurable Indicators	Progress and Achievements April 2004-Mar 2005	Actions required/planned for next period
<ul> <li>in resources to achieve</li> <li>The conservation of biological</li> <li>The sustainable use of its complexity</li> </ul>	-		countries rich in biodiversity but poor
Purpose			
To enable conservation management for the Peruvian population of the Andean guanaco.	The production of the management plan (at latest by the end of Year 3).	D-loop sequences had been produced for 4 populations and microsatellite analysis begun.	Complete other 2 population sequences and complete microsatellites for the remaining 6 populations.
To build capacity in conservation genetics in Peru.	The successful training two Peruvian scientists in conservation biology.	JR completed final year module in 2003.	KY to attend MWB final yr conservation module and get diploma and have intensive lab training
To train a cohort of Peruvian scientists in conservation biology and population viability analysis.	The courses having been successfully held and the trainees having earned their diplomas.	KY and JR completed the Peru course with diplomas and 13 Peruvian scientists also completed	Run second course in lima.
To carry out a Population Viability Assessment.	The production of a risk assessment for the guanaco. To have held the workshop		Preparation for Risk assessment to be conducted in next period

Outputs			
The production of six management plans INRENA can use to guide guanaco conservation.	The plans themselves should be easily translated into specific action.	To be completed in year 3.	Analysis is ongoing
Two scientists who can produce genetic data, analyse it and write scientific papers and management plans.	The scientists' increased knowledge and hands-on capability at conservation genetics should be verified.	JR was trained in automated sequencing and mitochondrial data analysis.	KY to be trained in genotyping and mtDNA analysis in Cardiff JR applying knowledge I Lima lab
Two training courses in conservation biology.	The students should be able to pass an exam at the end of their course or demonstrate increased knowledge.	Held June-July 2004 – all students got diplomas and were examined	Preparation for next course
A full population viaibility assessment.	The PVA can run successfully and provide useful indicators of specific threats and solutions for populations.	To be held in year 4.	Complete genetic lab work and analysis and ensure all relevant data gathered for PHVA

Note: Please do NOT expand rows to include activities since their completion and outcomes should be reported under the column on progress and achievements at output and purpose levels.