

DAVIS EXPEDITION FUND

REPORT ON EXPEDITION / PROJECT

Expedition/Project Title: Comparative phylogeographic study of western Amazonian species

Travel Dates: 1st April to 9th June 2011

Location: Bolivia and Peru

Group Members: Euridice Honorio

Aims: To improve the sampling of widespread species in western Amazonia across their southern ranges

Comparative phylogeographic study of western Amazonian species

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Summary:

This report summarizes a fieldtrip to Bolivia and Peru carried out from 1 April to 9 June 2011. The aim of this fieldtrip was to collect leaf material of southern populations of seven tree species that belong to the families Caricaceae (*Jacaratia digitata*), Moraceae (*Ficus insipida*, *Clarisia biflora*, *Poulsenia armata*), Myristicaceae (*Otoba glycyarpa*), Puntrajivaceae (*Drypetes amazonica*), and Arecaceae (*Iriarteia deltoidea*). A total of 328 samples in silica gel for DNA analysis and 47 samples for herbarium specimens were collected.

Introduction:

Chloroplast DNA (cpDNA) is increasingly used in phylogeographic studies for inferring genealogical histories of species such as population expansion and contraction, migration and colonization of new habitats through seeds (Avice, 2009). Even though the use of cpDNA is increasing in South America, only a few phylogeographic studies of neotropical plants have been published (e.g. Dexter, 2008; Dick *et al.*, 2007; 2008). Therefore, the world's most species-rich forests, located in western Amazonia, remain un-studied.

Western Amazonia lies on the eastern site of the Andes, and is characterized by strong edaphic, climatic and floristic gradients (Honorio *et al.*, 2009; Quesada *et al.*, 2009). This region contains the highest records of plant species richness in the world. Little is known about past vegetation dynamics in this region, but several past historical events have been identified that might have had an effect on the vegetation such as marine incursions (35 Ma; Hoorn & Vonhof, 2006), the uplift of the Andes (40-11 Ma; Graham, 2009), the formation of the Amazon River (11 Ma; Figueiredo *et al.*, 2009), and climatic oscillations during the Holocene (Hansen *et al.*, 1994). This PhD project aims to carry out a comparative phylogeographic study focusing on sampling the whole range of the geographical distribution of several species across western Amazonia to answer two questions: 1) Which colonization strategy have species used to achieve their widespread distributions in western Amazonia?, and 2) Has seed dispersal been a limiting factor for the geographical distribution of widespread species?

Preliminary results of my current PhD research suggest that the chloroplast DNA can be useful for inferring phylogeographic patterns of seed dispersal in western Amazonia. The most variable of seven cpDNA regions tested was *trnH-psbA* that showed a high number of potentially informative characters in most of the study species. In 2010, this region has been sequenced and amplified in 300 samples that belong to 5 species (*J. digitata*, *F. insipida*, *P. armata*, *C. biflora*, *O. glycyarpa*) collected across Ecuador and Peru. The haplotype networks suggest that recent expansion seems to have occurred in populations of central and southern Peru. High genetic differentiation is observed among populations in Ecuador and in northern Peru, which suggests that these populations have been more isolated.

Because phylogeographic studies require a sampling strategy that covers the entire range of the study species, a two-month fieldtrip to Bolivia and Peru was carried out to sample the southern geographical distribution of my target species to study the effect of ecological and historical factors on their distribution. The results of this fieldtrip are summarized below.

Methods:

Study area

Nine sites were visited in Bolivia and Peru near the eastern slope of the Andes (Fig. 1). In western Amazonia, precipitation regime shows long dry seasons in the south (>5 months with less than 100 mm/month) and more humid environments in the north (<2 months of dry season). I visited mainly sites in the Amazonian lowland rainforest (<600 m.a.s.l.) and only one site (Kenia) that was related to the seasonally deciduous Chiquitano forest. A few localities were also located in premontane forest such as Oriente Nuevo and Huampal in Peru which are >1000 m.a.s.l.



Figure 1. Sites visited during field trip in Peru and Bolivia, South America

Plant collection:

In each site at least 10 individuals of *Jacaratia digitata* (Caricaceae), *Ficus insipida*, *Poulsenia armata*, *Clarisia biflora* (Moraceae), *Otoba glycyarpa/parvifolia* (Myristicaceae), *Drypetes amazonica* (Puntrajivaceae), and *Iriartea deltoidea* (Arecaceae) were collected. To avoid sampling closely related individuals (e.g. offspring of the same parents), trees of at least 10 cm diameter, of different diameter sizes (and presumably ages), and separated by at least 40 m were chosen. Leaf samples of each selected tree was collected and dried in silica gel and the geographical location of the individuals were recorded using a GPS. Herbarium material was taken of each species at each site to be able to check species identification. Congenerics (other species of the same genus) were also collected if they were present in the sites.

Results:

Plant species:

Seven tree species were identified and collected in the field (Figure 2). These species represent the major phylogenetic clades of Angiosperms – Magnoliids, Commelinids, Fabids, and Malvids (Bremer *et al.*, 2009). *Otoba parvifolia* (Myristicaceae) and *Iriartea deltoidea* (Arecaceae) are distributed from Nicaragua in Central America south to Bolivia. In South America, they are restricted to western Amazonia, and grow in lowland and premontane rain forest up to 1500 m.a.s.l. (Henderson, 1990). While *I. deltoidea* is monospecific, the genus *Otoba* contains nine species (Gentry, 1979). The next three species belong to the family Moraceae, *Clarisia biflora*, *Poulsenia armata* and *Ficus insipida*, and are distributed from Mexico through the Andean region to Bolivia (Berg, 2001). They grow mainly on fertile soils and along streams in the lowland forest (Gentry, 1992). *Clarisia* has three species, *C. biflora*, *C. iliciflora* and the widespread species of *C. racemosa* and *Poulsenia* has two species, *P. armata* and an undescribed species from French Guyana. The genus *Ficus* comprises many species but only a few are trees (Berg, 2001). *Ficus insipida* consists of two subspecies, the widespread subspecies *insipida* and the subspecies *scabra* from French Guyana (Berg, pers.comm.). *Drypetes amazonica* is restricted to western Amazonia, and grow in lowland and premontane rain forest up to 1500 m.a.s.l. *Drypetes* is a pantropical genus and contains several species in Africa and Malasia, but only 3 species have been described in South America. *Jacaratia digitata* (Caricaceae) is restricted to western Amazonia from Ecuador south to Bolivia. This grows in the lowland rain forest on fertile rich soils (Gentry, 1979) and belongs to a genus of six species. The seeds of these species are dispersed by animals such as birds, monkeys, bats and fish (Zona & Henderson, 1989; Aide & Rivera, 1998; Zamora & Montagnini, 2007; Swamy, 2008).

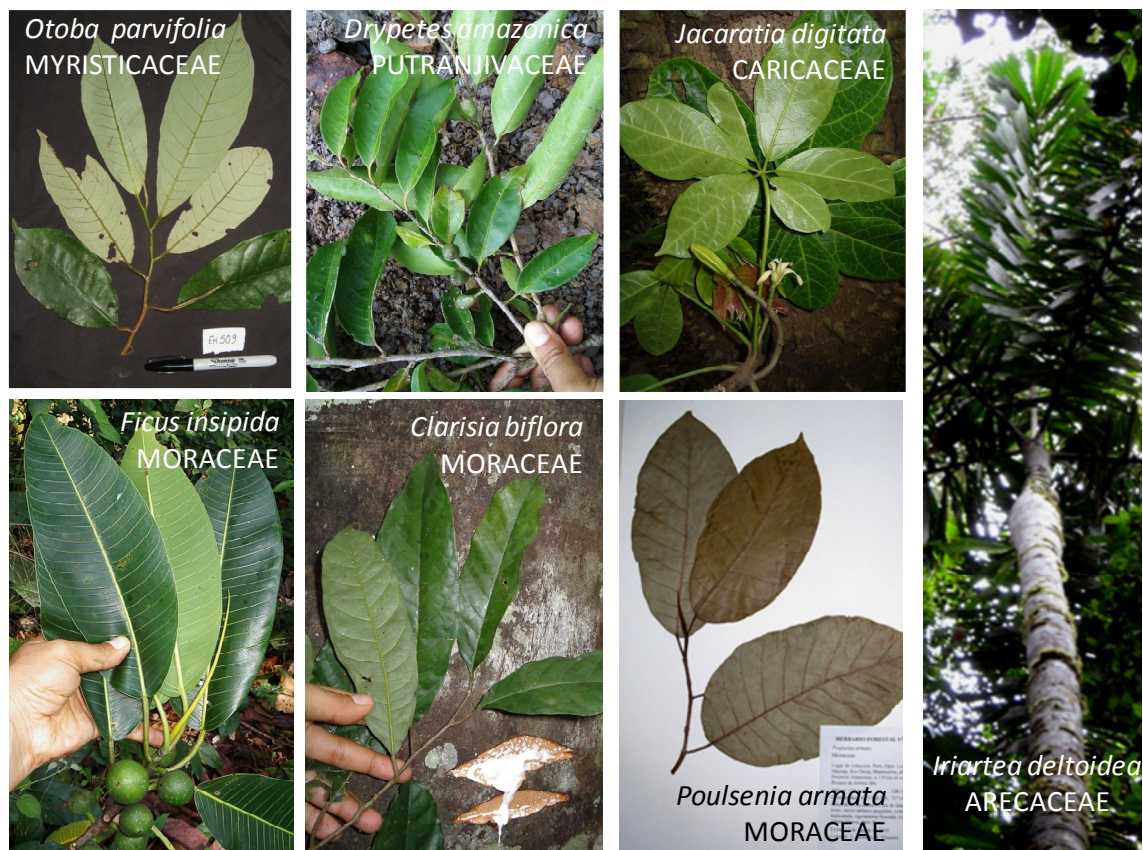


Figure 2. Seven tree species collected in western Amazonia

Plant collection:

A total of 328 samples in silica gel and 47 samples for herbarium vouchers were collected in the field. The material in silica gel will be used for DNA extraction and sequencing of a cpDNA region (*psbA-trnH*) at the Royal Botanic Garden Edinburgh. Herbarium specimens were deposited at the Noel Kempff Mercado Natural History Museum in Santa Cruz and the 'Referencia Amazonica' Herbarium in Cobija, Bolivia and the MOL Forestry Herbarium in Lima, Peru.

In Bolivia, most of the species were found at each site (e.g. *J. digitata*, *C. biflora*, *F. insipida*, and *I. deltoidea*, Table 1). Two species, *P. armata* and *O. glycyarpa*, were only collected in central and southern Bolivia, and *D. amazonica* was only found in the south (La Envidia). The best sites for plant collection were Maije, Sacta and La Envidia in Bolivia where most of the species occur with relatively high numbers of individuals. Overall, *P. armata*, *O. glycyarpa*, and *D. amazonica* were least commonly collected; this may be because these species often grow on seasonally flooded forests that we were unable to access because river levels were still high during the trip. *J. digitata* was only collected in Kenia, a site that is influenced by the seasonally deciduous Chiquitano forest. The other species were not found at this site, possibly because they are restricted to areas closer to the Andes.

In Peru, the collection focused on increasing the sampling of *C. biflora*, *P. armata* and *O. glycyarpa* (Table 1). Huampal and Tambopata were relatively easy sites for finding enough material of the study species; due to deforestation, Oriente Nuevo proved the most difficult site to sample.

Table 1. Number of samples collected in Peru and Bolivia. Sites are indicated with numbers as 1. Oriente Nuevo, 2. Huampal, 3. Tambopata (Peru), 4. Tahuamanu, 5. Abaroa, 6. Maije, 7. Sacta, 8. La Envidia, 9. Kenia (Bolivia)

Scientific name	1	2	3	4	5	6	7	8	9	TOTAL
<i>Jacaratia digitata</i>	0	0	7	10	10	10	10	10	10	67
<i>Clarisia biflora</i>	2	2	4	14	10	12	11	10	0	65
<i>Ficus insipid</i>	0	0	5	6	6	10	10	11	0	48
<i>Poulsenia armata</i>	9	8	0	0	0	5	10	10	0	42
<i>Otoba glycyarpa</i>	4	6	6	0	0	11	11	0	0	38
<i>Iriartea deltoidea</i>	0	0	0	12	5	10	10	0	0	37
<i>Drypetes amazonica</i>	0	0	2	0	0	0	0	10	0	12
Others	1	3	2	8	1	2	1	1	0	19
TOTAL	16	19	26	50	32	60	63	52	10	328

Collaboration and permits:

This fieldtrip was a great opportunity to collaborate with local researchers in Bolivia and Peru. Alejandro Araujo and Luzmila Arroyo from the Noel Kempff Mercado Natural History Museum in Santa Cruz were my main collaborators in Bolivia and Carlos Reynel from the La Molina Agrarian National University in Lima, Peru. Several other researchers were contacted to visit different sites such as Guido Vasquez and Julio Rojas from the 'Referencia Amazonica' Herbarium in Cobija, Stephan Beck and Narel Paniagua from the National Herbarium in La Paz, and Casimiro Mendoza from the Mayor de San Simon University in Cochabamba, Victor Landivar from the Noel Kempff Mercado Natural History Museum in Santa Cruz and Aniceto Daza from the La Molina Agrarian National University in Lima.

Finally, two research permits were obtained, one in Bolivia (SERNAP-DMA-CAR-1232/10) and one in Peru (Resolucion Directorial Nº 281-2010-AG-DGFFS-DGEFFS).

Financial statement:

The following table includes a summary of how the funding was spent during the fieldtrip carried out from 1 April to 9 June 2011 in Bolivia and Peru. A total of £ 3,740 was spent in transportation, accommodation, food, field assistants, and others. An amount of £ 3,500 was covered by the Davis Expedition fund and a remained of £ 240 was my personal contribution.

Table 2. Cost of fieldtrip carried out from 1 April to 9 June 2011 in Bolivia and Peru

EXPENSES	DETAIL	TOTAL (£)
Transportation	Leeds-Lima-Leeds	839.92
Transportation	6 sites in Bolivia	700.00
Transportation	3 sites in Peru	300.00
Accommodation	6 sites in Bolivia	320.00
Accommodation	3 sites in Peru	110.00
Food	6 sites in Bolivia	320.00
Food	3 sites in Peru	120.00
Field assistant	6 sites in Bolivia	600.00
Field assistant	3 sites in Peru	250.00
Equipment		180.00
TOTAL		3739.92

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