DAVIS EXPEDITION FUND

REPORT ON EXPEDITION / PROJECT

Expedition/Project Title:	Comparing the effects of different geological and ecological histories in shaping the genetic variability and population structure of populations of an ancient savannah plant species (*Senegalia mellifera (Vahl)*).
Travel Dates:	23rd September -25th October 2011
Location:	Namibia, Botswana, South Africa
Group Members:	Juan Carlos Ruiz-Guajardo & Samuel Thumbi
Aims:	Collection of leaf tissues from *Senegalia mellifera* for comparative phylogeographic and population genetic analysis between East and Southern Africa.

"Comparing the effects of different geological and ecological histories in shaping the genetic variability and population structure of populations of an ancient savannah plant species (*Senegalia mellifera (Vahl)*)".

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Summary:

This report summarizes an expedition to Namibia, Botswana and South Africa conducted from 20th Sept to 25th Oct 2011. The aim of the fieldwork was to collect leaf material of the important acacia tree *Senegalia mellifera* (Vahl), from several geographic localities across southern Africa. A total of 673 samples were collected from 24 different locations across Namibia (14), Botswana (8), and South Africa (1). Leaf tissues were collected fresh, and dried in silica gel to preserve DNA quality for future phylogeographic analyses. The samples and the database with geographical coordinates and populations have been deposited at Prof. Stone lab, IEB, University of Edinburgh.

Background:

The geological history of Africa has created complicated ecological gradients that constitute an excellent scenario for speciation and diversification. During the last 6 million years, severe climatic fluctuations have produced significant vegetation shifts that have resulted in the creation of isolated patches of habitat with unique ecological conditions that harbour several endemic species (Kingdon 1990). Examination of the present and historical distribution of endemic species can help improving our understanding of the forces driving speciation. A species' spatial distribution is largely dictated by a combination of past climatic shifts, historical changes in the distribution of communities, geographic features impeding migration, and by modern anthropogenic activities increasing fragmentation (Bobe 2006; Farwig et al. 2008; Kebede et al. 2007; Trauth et al. 2005; van Zinderen Bakker & Mercer 1986). A powerful way to unravel a species' evolutionary history is to use molecular markers to assess the distribution of genetic variability, allelic richness, and measure connectivity among existent populations via migration rates. Examination of population genetic structure and genetic variability allows identification of populations of low diversity and significant barriers to gene flow, so guiding the management of phylogenetically distinct populations of threatened or key species (Amos & Balmford 2001; Frankham et al. 2002; Sork et al. 1999). *Senegalia (Acacia) mellifera* is a common and widespread species, dominant in some regions, on rangelands and savannahs throughout western, eastern and southern Africa, the Sahel east of the Niger River, and the southern Arabian Peninsula (FAO 1983). A paper by Ruiz-Guajardo et al (2010) showed significant differentiation among

populations of *S. mellifera* in Kenya, with the Rift Valley constituting an effective barrier to dispersal. Despite its wide distribution and economical importance little is known about the genetic variability, and population genetic structure of this species outside elsewhere in Africa. The aim of our expedition was to collect samples of this important Acacia species across Southern Africa. The samples will be used to assess the spatial distribution of genetic variability in this species across southern Africa; allowing us to understand how differences in geological and ecological forces operating in East and southern Africa have shaped the evolutionary history of this ancient savannah species.

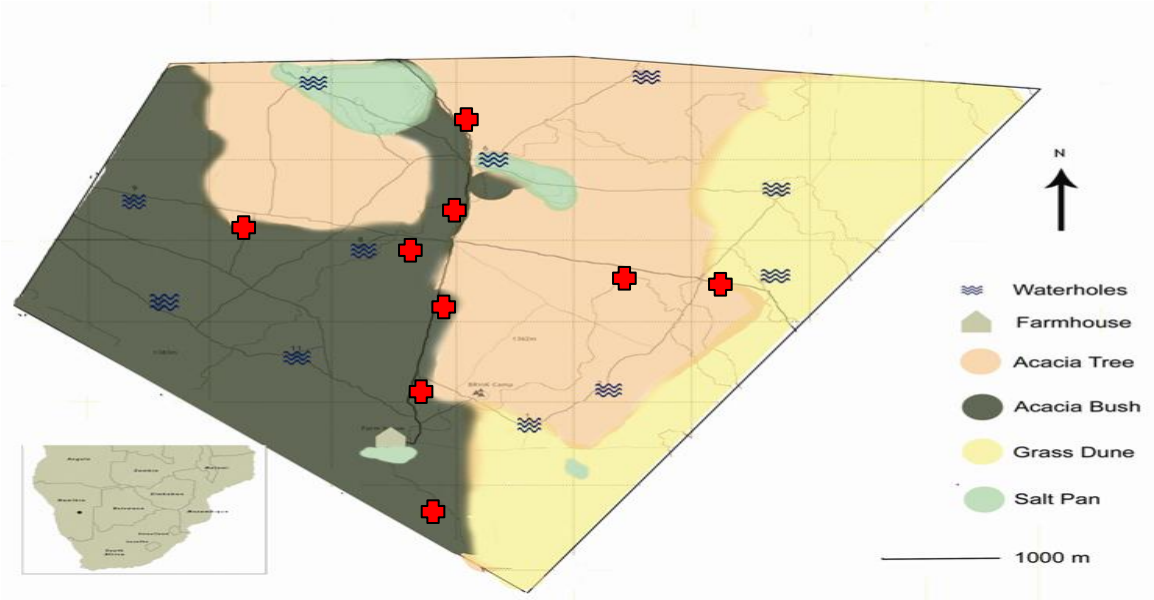
Expedition Account:

The expedition started on the 23rd of September when we flew to Cape Town, South Africa, where we collected essential equipment, and finalized the logistical details for the trip. Our sampling started in Namibia, then Botswana and finished in South Africa. Throughout the expedition we either stayed in youth hostels and guest houses, or camped if visiting remote bush areas. The sampling route followed the geographic distribution of *S. mellifera* in southern Africa published in (Van Wyk & Van Wyk, 1997). Overall we drove around 4700 km, and collected 673 samples from 24 locations across Namibia, Botswana and South Africa, making our collection the most comprehensive sampling for this species in Southern Africa (Table 1). At each locality, we targeted 30 individual trees; however this was not always possible due to low tree densities in some sites. For each tree we collected fresh leaves and a GPS reading. Leaf tissues were collected fresh and stored in silica gel within 12 hours of collection to preserve DNA quality.

NAMIBIA (24th Sep-1st Oct):

On the 24th of September we flew to Windhoek, Namibia where we met our collaborators from Biological Research in Kusikuz (BRinK), and sampled our first population (Industrial area Windhoek; Table 1). A total of 15 sampling sites were visited in Namibia (Table 1). Three collections were conducted enroute to Kuzikus, our main study area (Windhoek Junction, Dordabis, and Pond; Table 1). Kuzikus was chosen as the main study area due to its proximity to Windhoek (180 km South-East 23°14'S, E 18° 23'E), and its very high density of *S. mellifera.* Kuzikus is a restored part of the Namibian Kalahari with 100km^2 of *Acacia* savannah, bush-veldt, salt-pans and grass covered dunes supporting endemic wildlife (Fig 1). The sampling design followed for the collections in Namibia will allow us to investigate broad patterns of genetic diversity across Namibia. In addition, the 246 individuals from 9 populations, collected within Kuzikus will allow investigation of population genetic structure, and gene flow over fine geographical scales (Map 1). After our work in Kuzikus was finished, our collaborators drove us towards Hosea Kutako International Airport, collecting three more localities

on the way (Namibia 10, Silvia's Farm 1, and Silvia's Farm 2; Table 1). On the 1st of October we collected a 4x4 vehicle at Hosea Kutako Airport, and drove towards Botswana.



Map 1. Map of Kuzikus with sampling sites in red colour. Map taken from Brink (Biological Research at Kuzikus webpage (http://brink-namibia.com/kuzikus.php, accessed on 5th December 2011).

Figure 1. Sampling of *Senegalia mellifera* individuals at Kuzikus Kalahari desert in Namibia

BOTSWANA: (1st Oct-10th Oct)

We cross from Namibia into Botswana at the Buitepos/Mamuno border post. Overall we collected 215 individuals sampled from 7 sites across Botswana, and drove approximately 2000 km of main and secondary roads (Figure 2). Our first sampling locality was 5km after crossing the border in Mamuno (Table 1). We then headed North towards the Maun area, collecting samples at Ghanzi, and Maun Town (Table 1). We travelled North of Maun into the Okavango Delta, Moremi National Park seeking for *S. mellifera*, but unfortunately no trees were found in the vicinity of the Okavango River. According to the van Wyk & van Wyk, the study species could be found widely following a South, South-East direction from Maun to Gaborone the capital city. Hence, we followed this route stopping regularly to seek for suitable collection sites. Unfortunately, due to most of the land being included on a large scale veterinary experiment investigating foot and mouth disease control strategies, we could only collect samples at one locality on the way to Gaborone (Rakops, Table 1, Map 2). Once we reached the capital, we collected samples covering secondary alternative roads attempting to close the gaps in our sampling area, and ensuring a good collection representative of the genetic variability east of the country. We collected 3 sampling localities around the Gaborone area (Gaborone, Kanye, and Molepolole; Table 1, Map 2).

Map 2. Map of Botswana showing sampled localities as red triangles, and foot and mouth disease control experimental plots as green rectangles.

Figure 2. Common giraffe *(Giraffa camelopardalis)* in the surroundings of Moremi National Park, Okavango Delta, Botswana. Giraffe's favourite food consists of acacia leaves and other bushes.

Figure 3. Camping during the expedition allowed us to explore remote areas located far main towns

Figure 4. Crossing the Tropic of Capricorn enroute to South Africa, an iconic moment in the expedition

Figure 5. One of the multiple veterinary control check points running field experiments to investigate foot and mouth disease spread across Botswana.

SOUTH AFRICA (11th -25th Oct):

Senegalia mellifera in South Africa is restricted to the third most Northern part of the country. We aimed to drive back from the Botswana Pitsane back to Cape Town where we will travel back to the UK. According to van Wyk & van Wyk, we could have followed two potential sampling routes. The first alternative involved crossing the Kruger National Park, passing through isolated and remote areas, driving around 2000 km, but potentially finding *S. mellifera* only within the first 300 km. The second route involved driving a more direct route (~1600 km), covering around 600 km of the purported distribution of *S. mellifera*. Hence, to maximize our chances of encountering suitable patches populations of the study species, we decided to take the N12 road that crosses the country north to south. Recently, vast extensions of land surrounding the N12 north section have severely been modified by intensive agricultural practices involving irrigation and cattle projects (see Fig 9). This in combination with the encroaching that *mellifera* produces in over grazed areas, has led farmers to nearly completely extirpate this species, and as a consequence and despite our best efforts to find suitable sampling sites, we were only able to locate one population (Christiana, Table 1, Map 3). After our arrival to Cape Town, I met with Prof. David Ward, from Kwazulu Natal University who agreed to provide extra samples collected by his team across the country that will fulfil the gaps in our sampling of South African genotypes.

Map 3. Map of South Africa with Christiana as a red cross, the only population of *S. mellifera* that we were able to find between the border cross of Pitsane in south eastern Botswana, and Cape Town in SA.

Figure 6. Some road signs across South Africa. On the left the way to Edinburgh, a tiny village in the Mhala Northern province; and on the right the way to the famous diamond mining town of Kimberley.

Figure 7. Illustration of the highly modified landscape along the N12 between Pitsane and Kimberley. Most of the original *S. mellifera* savannah in this area has been modified into agricultural/cattle farmland through the use of intensive irrigation.

Closing statement:

Despite not being able to collect as many samples in Botswana and South Africa as originally planned, overall, the expedition was very successful. We managed to collect a very good number of samples across Southern Africa, drove more than 4600km, without facing any major road problems, and did not have any major difficulties with wildlife. Our sampling represents the most comprehensive collection of *S. mellifera* individuals in Southern Africa, and will allow examination of the population genetic variability and structure at two different geographical scales, a wide and a regional geographical scale. Analyses at a wide geographic scale will include all samples from Namibia, Botswana and South Africa, and will provide a good representation of the effect of geological history and historical gene flow within the area. Analyses at the regional scale will include only samples contained within Kuzikus, which will provide a finer resolution to understand the role of localised ecological gradients in shaping gene flow at short distances. The data generated from the samples collected will not only be original, and of high quality, but when combined with the data presented in Ruiz-Guajardo et al (2010), will constitute the first plant study to contrast Southern and East Africa, two regions high in diversity and with very different geological histories. The expedition was also successful in opening doors for future collaborations with institutions such as the University of Botswana, and Stellenbosch and Kwazulu Natal University in South Africa. The molecular work will be conducted in Prof Stone's laboratory in Edinburgh, and might constitute a good MSc/ Honours thesis project.

Acknowledgements:

We would like to thank Prof. Graham Stone and Dr. Richard Ennos for their support and guidance in planning this expedition. We sincerely thank The Davis' Expedition Fund, Edinburgh University for their invaluable financial support. We thank Prof. David Ward, Dr. Rouvey Roodt-Willding, and Dr. Gareth Hempson for their help with logistical support; and Johanna Reinhard from BRinK for her help organising and conducting fieldwork at Kuzikus. We thank Sylvia van Resburg from Astra Farm in Namibia, and Peterson Thumbi in Maun, Botswana for their wonderful hospitality.

Finances:

Table 2 presents a summary of the expenses incurred during our fieldtrip conducted from 20th September - 25th October 2011 through Namibia, Botswana and South Africa. A total of £6607.21 was spent in transportation, accommodation, food, field assistants, and others. An amount of £ 6,000 was covered by the Davis Expedition Fund, and the remaining £607 represent our personal contribution to the expedition.

Table 1. Sampling localities across the three countries visited, with UTM coordinates, and the number of samples collected at each geographic location (**N**).

Population	Country	UTM Northing	UTM Easting	N
Windhoek Industrial	Namibia	713950	7502438	27
Windhoek Junction	Namibia	740393	7506233	16
Dordabis	Namibia	777559	7460067	32
Pond	Namibia	207659	7436756	19
Kuzikus T088	Namibia	234666	7434768	31
Kuzikus 2	Namibia	233782	7433921	30
Kuzikus 3	Namibia	233594	7432712	16
Kuzikus 4	Namibia	234654	7431463	20
Kuzikus 5	Namibia	234477	7430440	30
Kuzikus 6	Namibia	234154	7428867	31
Kuzikus 7	Namibia	233965	7428729	30
Kuzikus 8	Namibia	235838	7431892	30
Kuzikus 9	Namibia	236832	7432345	30
Namibia 10	Namibia	205524	7502713	30
Sylvia's Farm 1	Namibia	205580	7502750	30
Sylvia's Farm 1	Namibia	198513	7520516	28
Buitepos Border	Botswana	404614	7537184	30
Ghanzi	Botswana	567134	7598086	29
Maun	Botswana	755551	7783202	35
Rakops	Botswana	231521	7669193	30
Gaborone	Botswana	386091	7669212	30
Kanye	Botswana	335889	7231969	31
Molepolole	Botswana	344587	7303024	30
Christiana	South Africa	290396	6889732	28
Total				**673**

Table 2. Summary of costs incurred during our expedition to Southern Africa

EXPENSES	DESCRIPTION	TOTAL (£)
Flights	Edinburgh-SA-Edinburgh	1778.48
Flights	Cape Town-Windhoek	300.00
Car Hire	Namibia-SA	1990.58
Petrol	4800 km	652.60
Accommodation	Namibia,Botswana,SA	695.55
Transportation	Regional	231.00
Permits	Border Fees, Research	65.00
Food		145.00
Equipment	Silical gel, Camping, Bags	240.00
Other		509.00
TOTAL		**6607.21**

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