JAMES RENNIE BEQUEST

REPORT ON EXPEDITION/PROJECT/CONFERENCE

Expedition/Project/ Conference Title: The Orchid Pollination Project 2010

Travel Dates: June – July 2010

Location: Chiapas, El Soconusco, Mexico

Group member(s): Laura Riggi, Rudi Verspoor, Andrew Wiles, Ailsa Burns, Melissa Lennartz-Walker

Aims: The main aim of this expedition was to investigate the abundance and richness of Orchid bees in various habitats to assess whether the lack of available pollinators could be a threat to orchids re integration in coffee plantations in the Soconusco region of Mexico. More specifically, the project was interested in looking at bee-orchid interaction through pollinia collection.

OUTCOME (not less than 300 words):-

See attached report.

Introduction

The Soconusco region (15°19' N and 92°44' W) covers 5.475 km^2, nearly 7% of the State of Chiapas. Situated at the extreme South East of the State, it extends from the coastal plains of the Pacific coast, to the mountainous hills of the Sierra Madre (Figure 1.). The Soconusco is a biologically diverse tropical region, nowadays mainly engaged in intensive, extensive and subsistence cultivation. In particular, large areas of low to mid elevation areas are dedicated entirely to the cultivation of coffee, which has resulted in the replacement of much native forest. An important aspect to the biodiversity of Soconusco is the huge variety of native orchid species, especially epiphytic orchids. However, the current trend to reduce shade cover in coffee plantations in the area has prompted concerns about the impact of this on orchids, leading to a program aimed at sustainably integrating natural orchid populations into coffee plantations.

Pollination ecology has been identified as an important aspect of orchid ecology. In Soconusco, many epiphytic orchids depend on Euglossine bees for sexual reproduction, when they transport their pollinia (clumps of pollen grains) from flower to flower. However, the pollination of orchids is rare and their remains little known about the pollination ecology of many species. Our study investigated the influence of habitat and environment on the presence and abundance of Euglossine bees as orchid pollinators, comparing forest fragments and coffee plantations. This contributed to a local project mapping specific pollinators to orchid species. This information in turn can be used to most effectively implement local orchid cultivation programs in coffee plantations.

Figure 1. Map of Mexico showing the position of the State of Chiapas.

For the study, we selected four field sites, based on diversity data from a previous study in the region. Sites were spread across an altitude range of 500 to 1,300m, and consisted of two coffee plantation, and two forest fragments, with "Edge" and "Centre", within each site.

Methods

At each site, three scented baits (photo 1), were attached to branches, at a height of 1.5m, and spaced by 4m. Baits were soaked in three different fragrances, known to be attractive to bees: Cineole, Eugenol and Methyl Cinnamate, and monitored for 4 hours, from 9-9.30am to 13.30pm. Sampling duration throughout the study was limited because of the time required to reach different sites and heavy afternoon rains. However, few bees were collected before 10.00am and bees are not active in heavy rain, so this not thought to have affected the results.

Photo 1. A scented bait surrounded by attracted male Euglossine bees.

Temperature, humidity, light, and wind speed were measured every 30 minutes using climate-metres. Bees were captured by hand-netting. The time of capture, type of bait, bee identity and presence of pollinaria was recorded for each capture. A sample of their leg was stored in alcohol for genetic analysis. Bees identified *in situ* were marked, using either acrylic paint or plastic numbered tags, and then released. These bees were then released. Bees that could not be identified were killed using ethanol, for later laboratory identification (less than 10%).

Results

Abundance and abiotic effects

In total, 1251 male bees from 14 species of 4 genera were collected during the 15 days of sampling. The most common species was *Euglossa tridentata* (45.63% of all specimens), followed by *E. viridissima* (18.86%). Analysis of variance (ANOVA) showed a significant effect of light intensity and relative humidity on the distribution of abundance of bees over all sites, (humidity: $F1/234=58.38$, $P<<0.001$; light intensity: $F1/234=4.27$, $P<0.05$, R-Sq(adj)=35.78%). Bee abundance was the highest when humidity was lowest, and when light intensity and temperature were highest. However, analysis of individual species shows that the trends were driven by *Euglossa tridentata*.

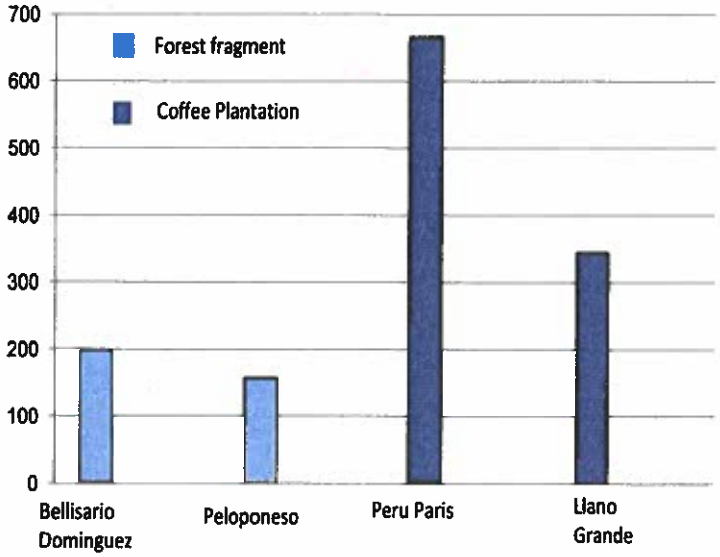


Figure 3: Histogram showing the total number of bees caught in each site

Habitat effect

Differences in bee abundance between coffee plantations and forest fragments were significant ($F_{1/234}=82.17$, $P<<0.00$, R-Sq(adj)= 25.65%), with most bees being caught in coffee plantations (Figure 2). This difference is primarily driven by Peru Paris site, which showed especially high abundances (N=669, 53.5% of bees caught). We therefore cannot conclude a significant habitat effect on bee abundance. However, Euglossine bees are present and abundant in coffee plantations.

Pollinia collection

63 (5%) of all bees captured were carrying one or several pollinia, from 8 orchid species, as well as 5 species which remain to be identified. The majority of bees carrying pollinia were found in coffee plantation sites. Species of pollinia from two endangered orchids were collected: *Kefersteinia tinschertiana* and *Cycnoches ventricosum*, from Peru Paris and Belisario Dominguez respectively. 18 *E. tridentata* individuals were caught in Peloponeso and Peru Paris, carrying pollinia from *Cycnoches ergotonianum*, for which very little data is available. Across the season, we found *E. viridissima* carried pollinia from 4 species, and *E. tridentata* from three, suggesting that bees can provide pollination services for multiple species of orchid.

Genetic studies

Our group is currently completing genetic analysis on the samples collected. The aim of this study is to challenge and confirm the use of morphological identification for Euglossine bees.

Discussion

Despite the time limitations of the project, our project made a significant contribution to knowledge of orchid pollination in the Soconusco. With regard to habitat, we found that orchid bees to be numerous and equally diverse in coffee plantations, when compared to forest fragments. The pollinia collections, the majority of which were from coffee plantations, also serve as evidence that bees in these areas are providing pollination services to multiple orchid species, some of which are considered threatened. The success of our collections is particularly notable due to our study being conducted during the rainy season when the majority of orchid species are not in flower, suggesting year round intensive sampling would yield informative results. Within coffee plantations we recorded a bee carrying the pollinia of

the rare orchid *K. tinschertiana,* suggesting that there are still remnant populations of orchids within cultivated areas. In combination these results support the idea of cultivating orchids within coffee plantations, as a viable system to promote long term and stable populations of orchid species.

Focussing on the abundance and diversity of Euglossine bees across sites, and their pollination behaviour, our study revealed that sites which are geographically relatively close can show differing dominant bee species, for example in comparing the Peru-Paris site with the Llano Grande site. However, bee communities were found, in general, to be similar across sites and habitats. Results from the collection of pollinia suggest that some bee species serve as pollen vectors for multiple orchid genera. In addition, the same orchid pollinia of some species can be carried by multiple species of bee. Continued work could investigate whether these species are equally competent in providing pollination services.

Our project has contributed to the understanding of orchid pollination in the Soconusco region, both with regard to species specific interactions between orchids and bees and also in exploring the general activity patterns and abundance of euglossine bee species. However, due to the short time frame of the project, there remains much work to be continued in order to further explore the subject of orchid pollination. Our results have been left with the leader of the orchid reintroduction programs where they can be used to best effect for orchid conservation in the Soconusco, and will provide a basis expanding studies using an intensive sampling approach.

Summary of group achievements

While our project had a strong focus on the scientific aims, it was also an important opportunity four personal and group development and provided a unique opportunity to explore independent work. The following summary provides a brief overview of the difficulties and achievements of our project.

During the project, our group faced some difficulties, both in planning and preparation, as well as throughout the field season. In 2009, our group was forced to cancel due to the swine flu situation in Mexico, due to funding being withdrawn. However, we remained resolved to complete the expedition the following summer. Shortly before our 2010 departure, one of our team members, Quentin Roland, had to withdraw from the field team due to unforeseen academic issues, resulting in a restructuring of group roles and responsibilities. Nonetheless, our arrival in Mexico went as planned. Within Mexico, our apartment was double booked resulting in an unexpected cost incurred from alternative accommodation. In the field, safety issues regarding animosity towards westerners in one field site resulted in a decision to cancel one visitation. However, in planning and preparation, our risk assessment facilitated our group in dealing with arising issues, resulting in the successful completion of the project.

In review of the project overall, we feel that the expedition was executed very successfully, both from the group and individual perspective. Our aims of investigating bee behaviour and pollination interactions have yielded preliminary results of immediate value to orchid conservation and pollination ecology, in addition to serving as a foundation upon which further studies can build upon. From the point of view of the individual, each of our team

members were able to gain a range of valuable skills throughout the project including; expedition planning, field medical skills, fundraising experience, and field skills. This experience will be invaluable in allowing us to contribute to both science and conservation programs in the future, as well as aiding us in our continued studies. In addition, our group were able to forge a relationship between Ecosur and the University of Edinburgh, which we hope will facilitate further collaboration on future projects and expeditions. Our group is also currently working on a more extensive report from which presentations and feedback can be provided.

Orchid conservation and management

Despite a lack of site and seasonal repeats due to time constraints, our results revealed some aspects of Euglossine bee responses to habitat quality and general activity patters preference. The results presented here suggest that coffee plantations may effectively support viable populations of orchid bees and the conservation status of these areas should be considered with great care. In general, it appears that pollinator availability is very variable in Soconusco; this question must be addressed for sustainable orchid production and conservation programmes. Some of the pollinators observed in this study coincide with other studies, but reports show clearly that orchids may be successfully pollinated by different pollinators in different areas, although possibly with reduced efficiency.

While this study shows that Euglossine bees are present and there is the potential for pollination, if orchid numbers are severely reduced, attention of available insects may be diverted towards new and richer sources of nectar, resins and fragrances. More research on orchid cultivation and re-integration within coffee plantations will help maintaining such fragile interaction between Euglossine and orchids.

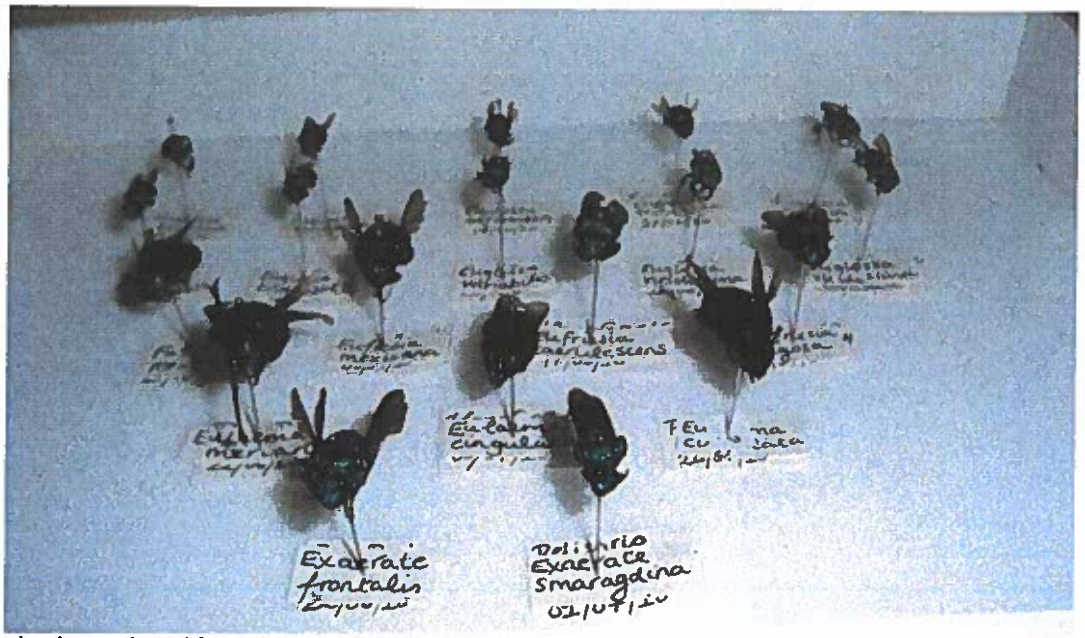
Acknowledgements

We would like to thank Dr. Anne Damon who helped us organizing this project since 2009 and whose help in the field and guidance have been invaluable. In addition we would like to thank the Institute of ECOSUR, Tapachula, Mexico, in particular Nelson Pérez Miguel, for helping us in the field, Guadelupe Nieto López, for identifying the pollinia samples, Dr. Daniel Sanchez, for his advice on genetic samples techniques, Eduardo Chamé Vázquez, for permission to transport the bees samples back to the UK, Vincenzo Bertolini, for his support, and Dr. Francisco Infante for his permission to work in ECOSUR. Many thanks also to Dr. Richard Ennos, Dr. Graham Stone and Juan Carlos Ruiz from the University of Edinburgh for supporting this project all along and for illuminating discussions. This project was supported by the Small Project Grant (Edinburgh University), the Royal Scottish Geographic Society, the Edinburgh University Travel Fund, the James Rennie Bequest, the Weir Fund, the Carnegie Expedition Fund, the Davis Fund (Edinburgh University), the John Rae Trust and the Explorers Club. Finally we would like to thank all our family members, friends and members of the Expedition society who encouraged and assisted this project.

Appendix - Photos

From left to right: Anne Damon identifying bees in the field; Melissa; Andrew; Andrew and Rudi working hard; Rudi and Laura entering data.

Field work. From left to right. Monitoring of vegetation in Peru-Paris (Ailsa and Rudi). Male Euglossa bees attracted to Cineole-scented bait. Catching of bees with plastic bags (Laura). Identification and marking of bees (Anne Damon and Andrew).



Euglossine bees pinned for collection and further identification.

APPENDIX

Number of male euglossine bees captured in four sites in southern Mexico during 15 field days between June and July 2010.

	Forest		Coffee		
Species	**Belisario Dominguez**	**Peloponeso**	**Peru-Paris**	**Llano Grande**	**Total**
Euglossa species ?	2	1	27	51	81
Euglossa tridentata	70	79	449	6	604
Euglossa variabilis	4	3	20	5	32
Euglossa atroveneta	1	0	5	2	8
Euglossa villosa	0	0	0	4	4
Euglossa townsendi	24	9	24	27	84
Euglossa crininota	6	5	21	22	54
Euglossa viridissima	23	38	64	87	212
Eulaema meriana	4	9	27	4	44
Eulaema cingulata	7	5	1	1	14
Exaerete frontalis	20	0	15	5	40
Exaerete smaragdina	13	0	9	0	22
Eufriesia mexicana	14	4	7	22	47
Eufriesia caerulescens	0	0	0	1	1
Eufriesia rugosa	0	4	0	0	4
Total	188	157	669	237	1251

Summary of average temperature, humidity and light intensity conditions in each site between Edge and Centre.

Site	**Edge**		
	Average Temperature (°C)	**Average Humidity (RH%)**	**Average Light intensity (Lux)**
BD	26,6	76,5	3025,2
PP	27,6	66,3	10799,3
LG	26,6	64,6	11492,1
PEL	24,5	76,6	6352,6
	Centre		
BD	25,9	81,6	1278,4
PP	27,3	67,9	7661,1
LG	23,5	77,2	1102,2
PEL	21,8	86,5	672,8

Table summarizing Pollinia data collected on at each location over the sampling period.

Location	Bee Species	Pollinia Species	Number of bees caught carrying pollinia from the same species
Belisario Dominguez	*Euglossa tridentata*	*Mieracycllium trinasatum*	*2*
		Un Identified	*1*
Llano Grande	*Euglossa Un Identified*	*Caudicula*	*1*
	Euglossa Un Identified	*Lycaste?*	*1*
	Euglossa Un Identified	*Stanhopea seccata*	*1*
	Eulaema meriana	*Cycnoches ventricosum*	*1*
	Euglossa viridissima	*Stanhopea seccata*	*4*
		Lycaste	*3*
	Eufresia mexicana	*Mormodes lineate*	*7*
	Euglossa townsendi	*Stanhopea seccata*	*3*
	Euglossa crininota	*Stanhopea seccata and Trichocentrum candidum*	*1*
	Euglossa variabilis	*Trichocentrum candidum*	*1*
Peloponeso	*Euglossa tridentata*	*Cycnoches ergotonianum*	*5*
		Stanhopea seccata	*1*
		Un Identified	*2*
	Euglossa viridissima	*Stanhopea seccata*	*1*
		Trichocentrum candidum	*2*
Peru-Paris	*Euglossa crininota*	*Cycnoches*	*2*
	Euglossa tridentata	*Cycnoches ergotonianum*	*13*
		Stanhopea seccata and Keferstenia lactea	*1*
		Stanhopea seccata	*4*
		Cycnoches X 2	*1*
		Un Identified	*1*
	Eulaema meriana	*Lycaste or Ornithocephalus tripterus*	*1*
	Euglossa townsendi	*Stanhopea seccata*	*1*
		Un Identified	*1*
	Euglossa variabilis	*Trichocentrum candidum*	*1*
	Euglossa Un Identified	*Un Identified*	*1*