DAVIS EXPEDITION FUND

REPORT ON EXPEDITION / PROJECT

Expedition/Project Title:	Chilean Expedition. *Genetics and Conservation of the Southern South American Conifers*
Travel Dates:	5th February to 10th May 2017
Location:	Central South of Chile, From the Maule to Aysén Region.
Group Members:	Mauricio Cano, Martin Gardner, Tom Christian and Peter Baxter.
Aims:	To collect DNA material, herbarium specimens, and seeds of the Chilean conifers.

Outcome (not less than 300 words):-

Royal Botanic Garden Edinburgh

2017

Chilean Expedition

Genetics and Conservation of Southern South American Conifers

THE UNIVERSITY of EDINBURGH

Mauricio Cano Niklitschek

Abstract

My fieldwork took place in Chile from the Maule to Aysén Region (35º-44ºS), comprising a three-month expedition. I focused on collecting DNA material, herbarium specimens, seeds and information from the field; patterns of regeneration and conservation pressures in four emblematic endemic conifers from South America: *Fitzroya cupressoides* (Cupressaceae), *Prumnopitys andina*, *Podocarpus salignus* and *Saxegothaea conspicua*, all members of the Podocarpaceae.

I covered a total of 42 populations encompassing the entire native range of each species. I gathered 25 DNA samples from most of the populations. Seeds were collected from most populations of *P. andina* and some northern populations of *P. salignus*, missing the southern ones due to seed availability. No seed production of *Fitzroya* and no seed dispersal of *Saxegothaea* was found, however, this latter species showed a huge immature seed production which will be ready for collecting next year (January-March).

The following field work summary report will provide an overview of the main achievements, the information that has been taken from the field and conclusions.

Introduction

Globally there are around 627 species of conifers, from eight families and 70 genera (Farjon 2008). Although the largest abundance of individual conifers is in the great boreal forests of Eurasia and North America, species diversity of the Southern hemisphere is higher in proportion to the available land area (Farjon 2008). In Chile, these southern hemisphere conifer species represent an iconic element of the flora. There are nine species representing 3/8 extant conifer families (Araucariaceae, Cupressaceae, Podocarpaceae) and eight genera, with all species and four genera (*Austrocedrus*, *Pilgerodendron*, *Saxegothaea* and *Fitzroya*) restricted to southern South America.

The Chilean conifers are concentrated in the Chilean Temperate Rainforest, one of the world's plant biodiversity hotspots, containing around 5000 species with almost 50% levels of endemism. Although some species of Chilean conifer such as *Araucaria araucana* (Monkey Puzzle) have been subject to intensive research, most have not. Knowledge gaps on their basic biology represents a limitation in the development of effective conservation strategies. This is a pressing challenge given extensive threats to plant biodiversity in the region (harvesting, climate change, pathogens, expanding plantation forestry and agriculture, and natural and human induced fires).

The aim of this project is to assess patterns of regeneration, population connectivity/differentiation, and conservation pressures in four emblematic endemic conifers from South America each with a restricted area of distribution. The study species are *Fitzroya cupressoides* (Cupressaceae); *Prumnopitys andina*, *Podocarpus salignus* and *Saxegothaea conspicua* (all Podocarpaceae). The motivation for the work is to understand the biology of individuals, populations and species and to develop conservation strategies in the context of emerging threats.

Area of study

The expedition took place in Chile from the Maule to Aysén Region (35º-44ºS). The longest distribution of the species involve in this project belongs to *S. conspicua*, which has a distribution from the latitude 35º to 46ºS. The other two Podocarpaceae; *P. andina* and *P salignus* present a central south distribution in Chile. *P. salignus* along both the Andean and coastal mountain range of Chile from 35° to 40°S. *P. andina* is in the Andes mountain from 35º to 39º S showing also a single population on the eastern slopes of the coastal range in the Araucanía Region. The Cupressaceae species (*F. cupressoides*) has a Southern distribution from 39º to 43ºS in Chile.

Figure 1. South America map. Red circle showing the conifers distribution in Chile.

Participants: *Mauricio Cano PhD student, University of Edinburgh and Botanical Garden of Edinburgh; Martin Gardner, Royal Botanic Garden of Edinburgh; Tom Christian Royal Botanic Garden of Edinburgh; Peter Baxter Benmore Botanic Garden*

Chilean Field assistant: *Fernando Bustos, forestry engineer; Alberto Nino, seed collector, Nicolas Lavandero MSc Royal Botanic Garden of Edinburgh, and Paloma C, local assistant, Reinhard Fitzcher; MSc in forestry ecology.*

Itinerary

Table 1. Schedule Chilean Expedition

TIME	ACTIVITY	AREA/POPULATION(S)	SPECIES TO COLLECT
6 February	Arrival to Chile from Edinburgh		
7- 13 February	Materials gathering, renting a car, food provision, meeting with assistant, coordination		
14 February	Arrival to San Clemente, Talca		
14-23 February	Collecting day, covering 8 populations;	Corral de Salas, Hornillos, Los Punquios, Puente Atacalco, New logging area, Fundo los Ciervos and Rio Quenco.	*P. andina, P. salignus, S. conspicua*
23-28 February	Collecting day, covering 6 populations; .	Angol, Nahuelbuta, Termas de Pemehue, RN Malleco, Victoria, Puento Loncotripan	*P. andina, P. salignus, S. conspicua*
1-3 March	Collecting day, covering 2 populations,	Cunco, Conguillio	*P.andina, , S. conspicua*
4-5 March	Collecting day, covering 2 populations	Reiglioni, Huerquehue.	*P.andina, , S. conspicua*
5 March	Computer work.	Coñaripe	
6-8 March	International workshop (Mycorrhizal symbiosis in the Southern Cone of South America)	Valdivia	
9-13	Collecting in Huinay	Huinay	*S. conspicua, Fitzroya*
14-25 March	Collecting	Austral region (Patagonia)	*S. conspicua, Fitzroya*
26-29 March	Computer work	Puerto Montt	
30 March-5 April	Collecting day, covering 5 populations,	Fundo Chahuilco, Cordillera pelada, RN Ñahuel ñadi, Fundo Chaquihue	*S. conspicua, Fitzroya*
6- 10 April	Collecting day, covering 4 populations,	Lanco, Los Lagos, Llancahue, Oncol.	*S. conspicua, P. salignus*
11 April	Computer work	Puerto Montt	
12-18April	Collecting day, covering 1 population	Valdivia region	*F. cupressoides, S. conspicua*
19-24 April	Break	Valdivia	
24-30 April	Collecting missing populations in the north of Chile, Maule region	Araucania and Bio Bio región	*P. salignus, P. andina*
1 May	Return to Valdivia		
2 May	Break		
3-9 May	Computer work, seed cleaning, samples checking	Valdivia	
10 May	Return to Edinburgh		

Data recorded

DNA samples

All populations previously proposed for the field work were covered, except for the northern coast population of *P. andina* which due to the catastrophic fire in Chile last summer (February 2017) made it impossible to visit the area. The northern Andean population of *S. conspicua* also could not be covered due to geographical barriers which made it difficult to access the population.

Number of DNA samples and populations covered

10 populations per species were covered; excluding *S. conspicua*, where a total of 12 populations were covered. A total of 25 DNA samples per population were collected. The images below show the population distribution for each species where the DNA material was collected, plus the estimated sample area covered.

P. andina

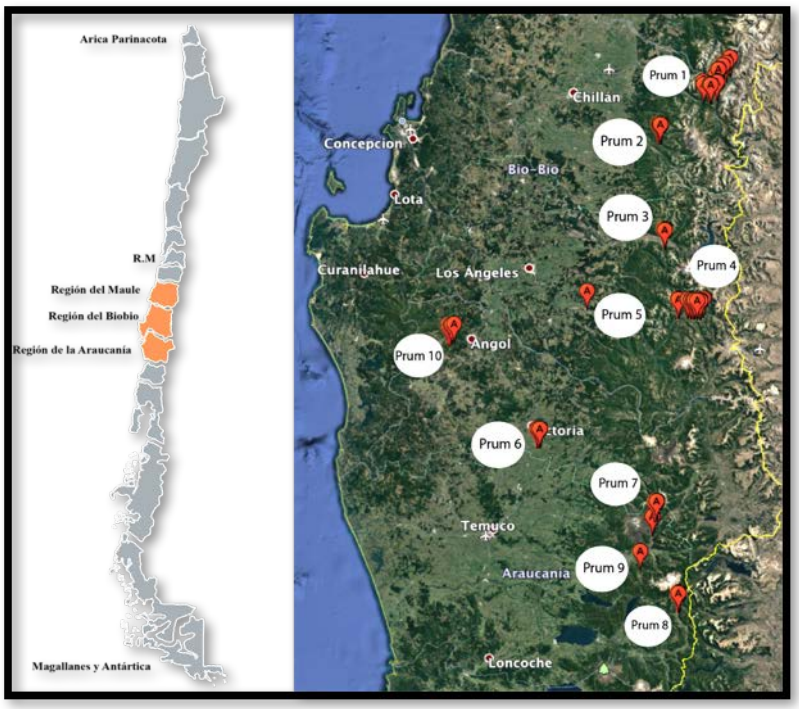

Figure 2. Populations of P. andina covered in the fieldwork.

***Locations:**: Prum 1: Corral de Salas; Prum 2: Los Punquios; Prum 3: Los Lleuques; Prum 4: Laguna del Laja; Prum 5: Trapa-Trapa; Prum 6 Pua; Prum 7: Conguillio; Prum 8: Reigolil; Prum 9: Nassampulli; Prum 10: Nahuelbuta.*

Sample Area per population of P. andina (ha)

Prum1	*Prum2*	*Prum3*	*Prum4*	*Prum5*	*Prum6*	*Prum7*	*Prum8*	*Prum9*	*Prum10*
3.27	413	0.92	3.1	468	16.11	46.4	2.28	13.2	28.9

P. salignus

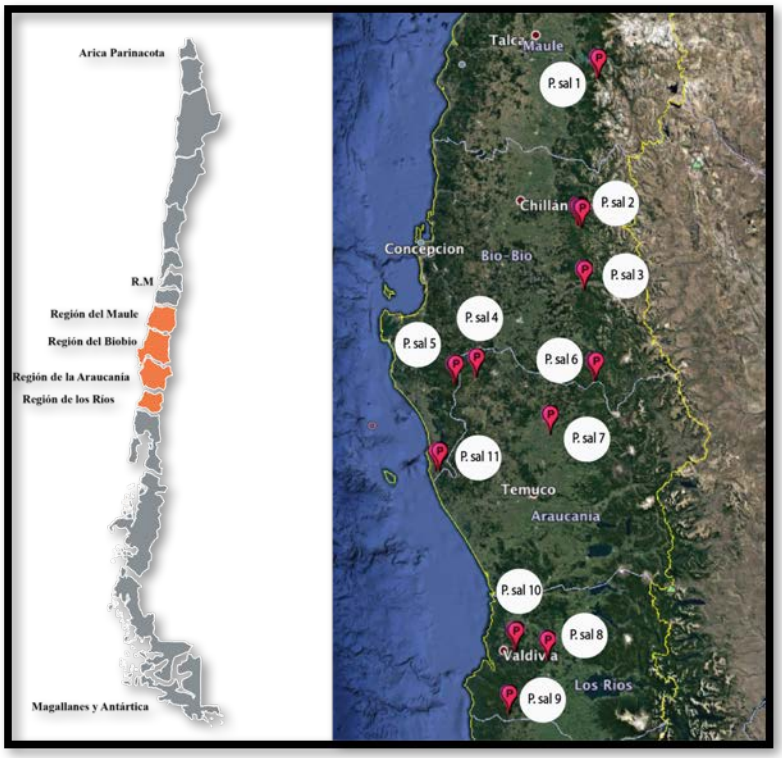

Figure 3. Populations of P. salignus covered in the fieldwork. Populations 4 and 5belong to a single population.

Locations. *Sal 1: Hornillos; Sal 2: Ñuble; Sal 3: Altos de Antuco; Sal 4-5: Nahuelbuta; Sal 6: Malleco; Sal 7: Pua; Sal 8: Reumén; Sal 9: Llancacura; Sal 10: Llancahue.*

Sample Area per population of P. salignus (ha)

Sal1	*Sal2*	*Sal3*	*Sal 4_5*	*Sal 6*	*Sal 7*	*Sal 8*	*Sal 9*	*Sal 10*	*Sal 11*
4.74	59.6	0.7	2.63	1.29	0.27	3.66	3.71	3.39	10

S. conspicua

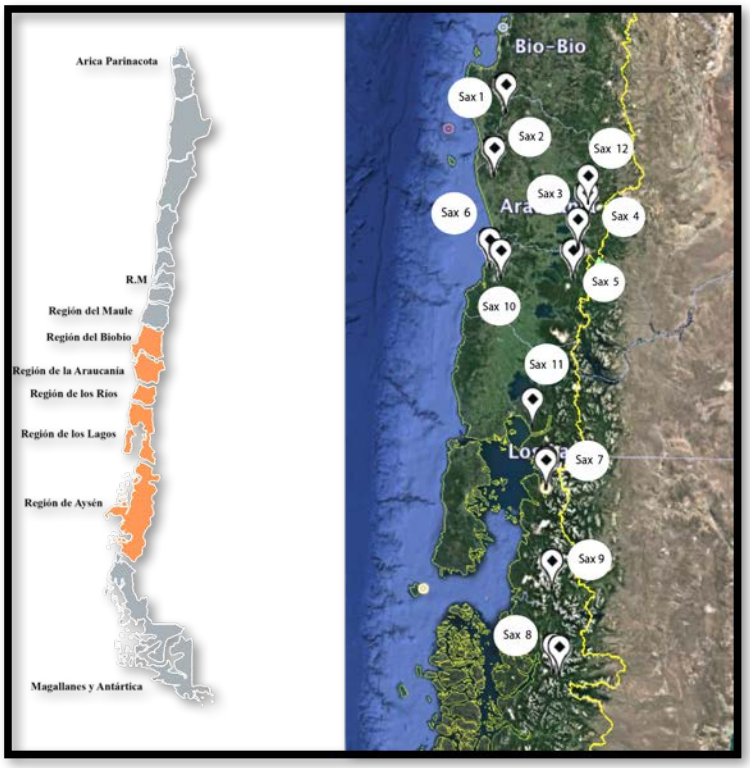

Figure 4. Populations of S. conspicua covered in the fieldwork.

***Locations**. Sax 1: Nahuelbuta; Sax 2 Villas las Araucarias; Sax 3 Huerquehue; Sax 4: N.P Villarrica; Sax 5: Huilo-Huilo; Sax 6: Oncol; Sax7: Huinay; Sax 8: Rio Cisne; Sax 9: Villa Santa Lucia; Sax 10: Llancahue; Sax 11: Lenca; Sax 12: Nassampulli.*

Sample Area per population of S. conspicua (ha)

Sax1	***Sax2***	***Sax3***	***Sax4***	***Sax5***	***Sax6***	***Sax7***	***Sax8***	***Sax9***	***Sax10***	***Sax11***	***Sax12***
9.45	11.6	4.1	14.3	23.7	3.15	none	19.94	19	0.59	37.7	5.1

F. cupressoides

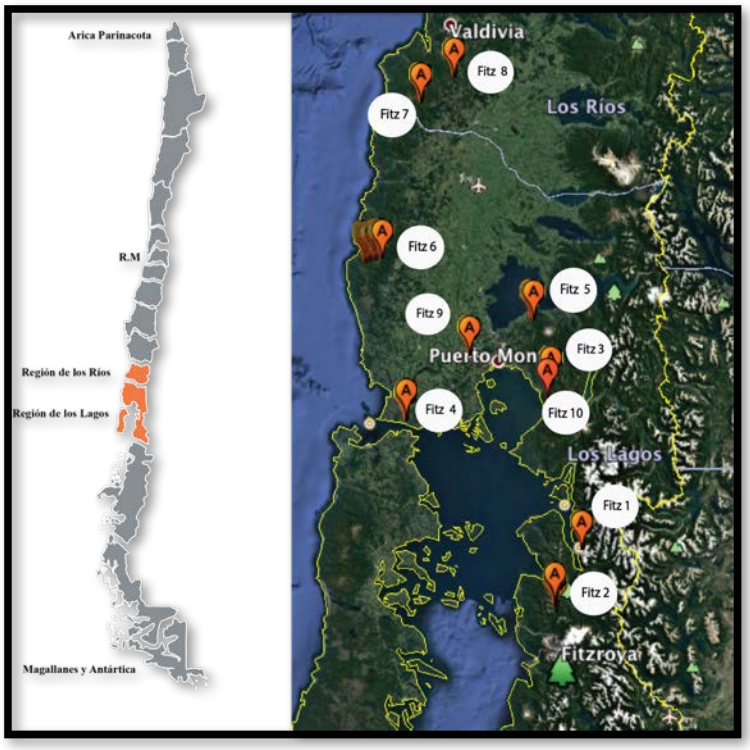

Figure 5. Populations of F. cupressoides covered in the fieldwork. The green pine icon shows the southern population which it was not covered.

Locations. Fitz 1: Huinay; Fitz 2: Caleta Gonzalo; Fitz 3: Alerce andino; Fitz 4: Astillero; Fitz 5: Fundo Rio Pescado; Fitz 6: Cordillera pelada; Fitz 7: Alerce costero; Fitz 8: Tres Chiflones; Fitz 9: Fundo Nuñez; Fitz 10: Lenca.

Sample Area per population of F. cupressoides (ha)

Fitz1	***Fitz2***	***Fitz3***	***Fitz4***	***Fitz5***	***Fitz6***	***Fitz7***	***Fitz8***	***Fitz9***	***Fitz10***
1.47	1.53	0.32	4	0.54	38.4	0.75	0.1	0.42	44

Seed material

Most populations of *P. andina* through its native range were producing seed. Seed production in *P. salignus* was only concentrated at the northern populations. In contrast, *F. cupressoides* did not show any signal of seed production except for a couple of small individuals at the Alerce costero population and one big individual in the central depression population (Fundo Nuñez). *S. conspicua* did has seed production, however, they were too immature to collect, giving the hope for collecting them next year. It was the same case with the *P. salignus* populations at the southern regions.

Number of seeds collected

The goal was to collect 10 individuals per population and 100 seeds per individual. The images below show a population map distribution per species where it was possible to collect seeds.

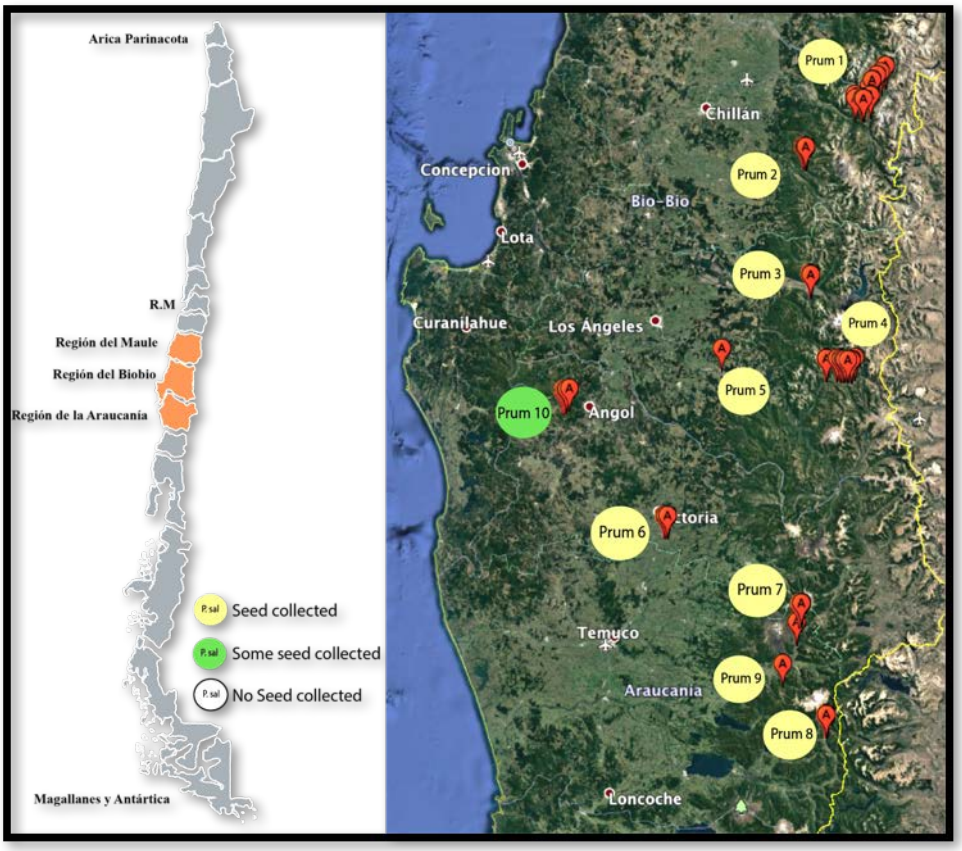

Figure 6. Populations of P. andina where it was possible to collect seed.

**Seed collected (yellow): 10 individuals, 100 seeds per individual.*

**Some seed collected (Green): 4 individuals, 100 seeds per individual.*

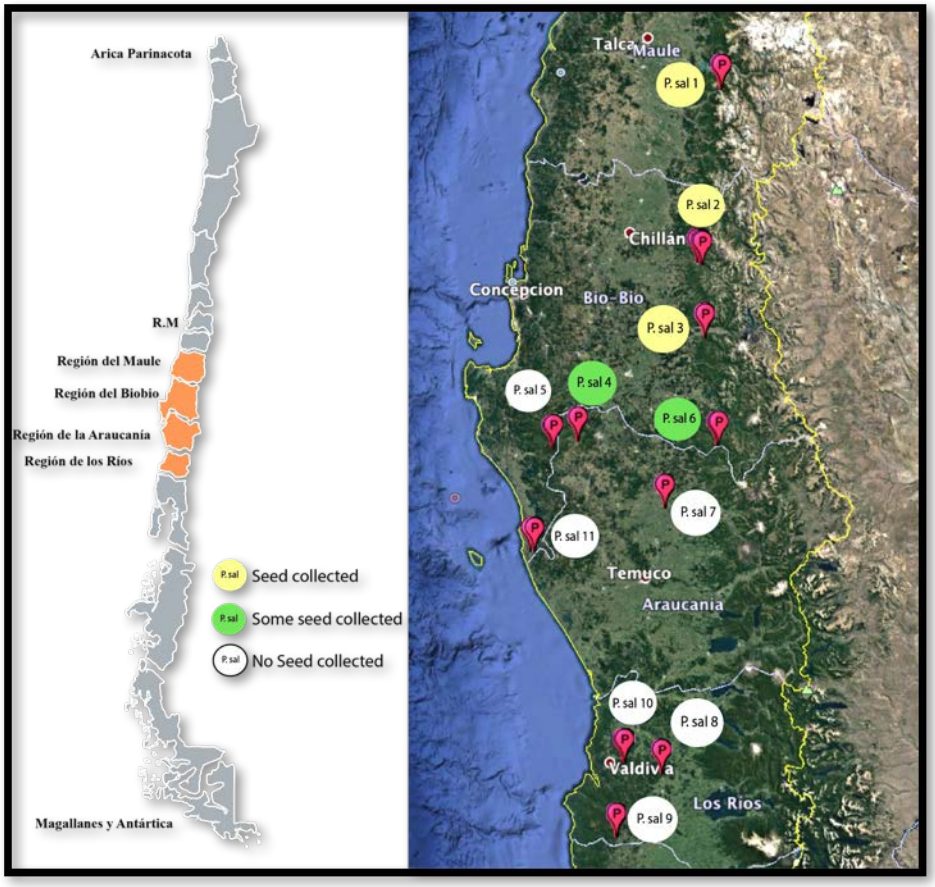

Figure 7. Populations of P. salignus where it was possible to collect seed.

Seed collected (yellow): 10 individuals, 100 seeds per individual.

Some seed collected (Green): **Pop 4; three individuals, 100 seeds per individual. **Pop 6**; four individuals, 100 seeds per individual*

Regeneration

Presence and absence of regeneration per population was recorded. However, it was very difficult to follow a proper methodology in the field due to the geographical barriers and in most cases, there was not enough time for collecting and recording all the samples/data per population.

Finally, the main methodology done in the fieldwork consisted of counting or estimating the regeneration (seedlings and saplings) under the trees where we were collecting seeds plus the places where we passed through.

Overall, *P. andina* populations showed most evidence of regeneration, with the Southern population at Nassampulli and Reigolil (Prum 9-8) being the most extensive ones, exhibiting a huge regeneration under canopy (table 2).

The small northern population at Corral de Salas (Prum1) did not show any evidence of regeneration at all. *P. salignus* also showed some regeneration in most populations, with the population at Altos de Antuco, sector el Colehual (Sal 3) being the one with the largest regeneration. *S. conspicua* showed two populations with lots of regeneration at population 2 and 3. *F cupressoides* did not show much regeneration in the field (table 2).

Table 2. shows the presence of regeneration per species/population and its maximum and minimum number of seedling and saplings observed.

Species/Population	1	2	3	4	5	6	7	8	9	10	11	12
P. andina	No	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	No		
Min	0	100	X	50	10	150	0	300	**1000**	0		
Max	0	150	X	100	20	200	0	500	**4000**	0		
P. salignus	X	Yes	Yes	Yes	X	Yes	Yes	Yes	No	Yes		
Min	X	X	**300**	10	X	100	100	50	0	25		
Max	X	X	**400**	30	X	200	200	100	0	50		
S. conspicua	No	Yes	Yes	Yes	X	No	No	No	X	No		
Min	0	100	**300**	20	X	0	0	0	X	0		
Max	0	200	**500**	30	X	0	0	0	X	0		
F. cupressoides	Yes	No	No	Yes	No	Yes	Yes	No	Yes			
Min	X	0	0	X	0	X	X	0	X			
Max	X	0	0	X	0	X	X	0	X			

***yes**: presence of regeneration. **x**: no information recorded. **no**: no evidence of regeneration. *Min: Minimum number of seedlings and samplings observed. *Max: Maximum number of seedlings and samplings observed.*

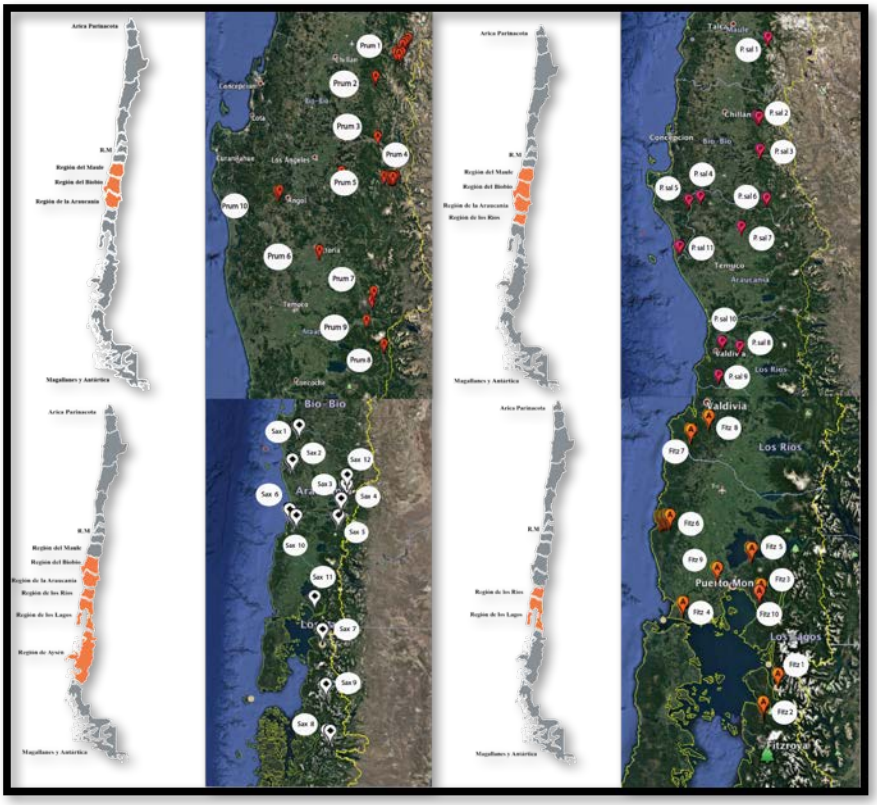

Figure 8. Population distribution of each species. top left, P. andina; top right, P. salignus; bottom left, S. conspicua; bottom right, F. cupressoides.

Conservation assessment

To identify the major threats for each species, I documented field information following the table below (table 2). The table displays a list of threats types extracted and modified from the IUCN *"Threats Classification Scheme (Version 3.2)"*. For each threat, the status (presence or absence) was recorded, its timing, its scope and its severity, assigning a respective value to each one (table 3). The values will soon be analysed and compared between population of each species.

Table 3. Threats types and their variables with values as an example.

Residential and comercial development	**Status**	**Timing**	**Scope option**	**Severity**
Housing and urban areas				
Commercial and industrial areas				
Tourism and recreation areas	1	0	1	0
Agriculture				
Annual and perennial non-timber crops				
Wood and Pulp plantations				
Livestock farming and rancing				
Energy production and mining				
Renewable energy				
Transportation and service corridors				
Roads and railroads				
Utility and service line				
Biological resource use				
Logging and wood harvesting				
Natural system modifications				
Fire and fires supression				
Dams and water maafement/use				
Other ecosystmen modification				
Invasive and other problematic species, genes and diseases				
Invasive non-native/alien species / diseases				
Problematic native species/diseases				
Introduced genetic material				
Problematic species/diseases of unknown origin				
Geological events				
Volcanoes	1	2	3	4
Earthquakes/tsumanis				
Avalanches/landslides				
Droughts				
Temperature extremes				
Storms and flooting				
Other impacts				
Other threat				
Impact	2	2	4	4

**Impact represent the sum of each variable*

Table 4. Variables and their values.

Values	***Timing option***
4	On going
3	In the past but now suspended and likely to return
2	Only in the future
1	Only in the past and unlikely to return
0	Unknown
Values	***Scope options***
3	Affects the whole population $> 90\%$
2	Affects the majority of the population 50-90%
1	Affect the minority of the population $< 50\%$
0	Unknown
Values	***Severity options***
4	Hight Impact
3	Medium Impact
2	Low impact
1	Unknown
0	No impact
	Status
1	**Prensent**
0	**Missing**

Conclusion

Generally speaking the expedition was quite successful. I got most DNA material, missing only a couple of populations which can be easily covered next year. Seed collecting was not easy in all populations, spending a lot of time, specially at the southern populations of *P. salignus* where most of the seeds were immature. It was not impossible to get seeds from *F. cupressoides* and will be even harder to get them next year. No evidence of green cones was found to suggest the possibility of a seed production for the next season. In contrast *S. conspicua* showed a huge seed production in most populations, which will be ready for collecting next year (2018).

Some limitations were experienced when collecting data from the field, especially collecting data about regeneration (seedlings and saplings). It was very difficult to follow the methodology proposed before the fieldwork, mainly due to time and to a lesser extent the geographical barrier shown in some areas. Besides these limitations, I could get an overview about the situation in most populations. This approach could definitely help for future methodologies.

The conservation assessment methodology made before the expedition was not difficult to implement in the fieldwork, however, the table used and its values (modified from the UICN) could be better adapted for collecting data.

Overall, the biggest limitations in collecting vegetative material from the species/populations was obtaining the collection permits. Even though, in the majority of the cases we got a permit for collecting, the area where we were allowed to collect did not represent the entire population of a species.

Acknowledgements

I would like to thanks to Davis Expedition Fund which made possible this expedition with its generous fund collaboration. I would also like to thanks the James and Eve Bennet Trust Expedition fund which also support this field work. I would like to mention and thanks the extraordinary collaboration of Martin Gardner, Tom Christian from the Botanical garden of Edinburgh and Peter Baxter from Benmore Botanical garden. From Chile, I had the collaboration of Fernando Bustos, Nino, Paloma, Nicolas Lavandero, Reinhard Fitzcher and all the local people who helped and made this expedition easier and friendly. Many thanks to my supervisors for their constant support.