



Conservation and vegetative propagation of forest genetic resources from Talares and Monte Blanco ecosystems in Argentina

Sebastián P. Galarco¹, Maite Romero Alves¹, Patricia Boeri², Luciano Roussy¹, Marina Adema¹, Blanca Villarreal¹, María V. Briones³, María de los A. Basiglio Cordal^{1,4}, Tatiana Cinquetti¹, Diego I. Ramilo¹ and Sandra E. Sharry^{1,2,3}

¹Wood Research Laboratory, School of Agriculture and Forestry Sciences, National University of La Plata, Buenos Aires, Argentina.

²Integrated Unit for the Innovation of the Agri-Food System of Northern Patagonia, National University of Rio Negro, Río Negro, Argentina.

³Biodiversity and Biotechnology Research Institute, National Council of Scientific and Technical Research, Buenos Aires, Argentina

⁴In vitro culture laboratory – CEAMSE (Ecological Coordination Metropolitan Area, State Society) . Villa Domínico, Buenos Aires, Argentina.

▫ Corresponding Author Email: ssharry@gmail.com

Abstract

Deforestation and forest degradation have led to widespread biodiversity and environmental losses in Argentina. The Talares and Monte Blanco forests are a relict vegetation type throughout Northeast of Buenos Aires province. These forests have suffered extensive deforestation and fragmentation. It is demonstrated that there is a low capacity of regeneration of the indigenous species after clearing, given that the growth of other previous species causes water deficit and lack of light (Arturi, 1997). That is why selection and successful propagation of native tree species are important for improving ecological restoration of these forests.

Buenos Aires province forests status

The native forests of Buenos Aires province are strictly confined to the coastal strip of Río de la Plata (coastal forests, locally "Talares" and "Monte Blanco") and to the western region (locally Caldén forests) (Parodi 1940). The use of forests for firewood was so intensive that the resources were exhausted at the beginning of the 19th century.

Currently there are only remnants or relicts of these forests, locally called "Talares," "Caldenal" and "Monte Blanco," as mentioned before. The native dry forest, dominated by *Celtis erenbergiana* (Ulmaceae) and *Scutia buxifolia* (Rhamnaceae), constitutes the main woodland community of the eastern plain called Pampa in Buenos Aires province (Parodi 1940; Cabrera 1976).

The Talares forests area is an environmental mosaic where thorny forests and woodland patches combine with humid grasslands and coastal communities of Río de La Plata. It is a xerophyte forest type structured by few tree species surrounded by a lower and moister soil matrix. The forest grows on highly calcareous parent materials, derived from sea transgressions and regressions in the Quaternary (Cabello and Arambarri 2002). The current status of conservation shows that the protection of these forests is considered a priority due to their high biodiversity and fragility. The unique characteristics of the Talares

led to the creation, in 1984, of the Biosphere Reserve “Parque Costero del Sur” (MAB-UNESCO), now considered a natural and cultural heritage site. This reserve consists of a group of natural interphases, with exceptionally rich flora and fauna. Forests dominated by *Celtis ehrenbergiana* are becoming susceptible to invasion by exotic species such as *Ligustrum lucidum* (privet). The tala tree is usually common in the riparian zone, although human activities limit its propagation. It is a valuable species for the survival of native animals as it serves as shelter, food source and bird nesting sites. Ribichich and Potomastro (1998) found that the dominant trees are *Scutia buxifolia* and *Celtis ehrenbergiana*. There is also present *Schinus polygamus*, which presents the bell-shaped structure of the pioneer species. Over 1200 m distant from the river, this species is co-dominated by *S. buxifolia* and by *C. ehrenbergiana* trees regenerated from stumps. The differences between the old-growth stands seem to be related to the gradients of soil texture and nutrient concentration, raising edaphic stress towards the river. The stress tolerance of *S. buxifolia*, and the aptitude of *S. polygamus* to survive in disturbed habitats seem to have prevented the post-logging survival of *C. ehrenbergiana*. Tala regenerated possibly due to a better competitive performance in a more favourable site. Ribichich and Potomastro (1998) recommend the restoration of the qualitative features and the control of privet. Goya et al. (1992) and Arturi and Goya (2004) have conducted several studies in order to protect the species. The reproductive system varies among the different species: *C. ehrenbergiana* is anemophilous and self-compatible. *S. buxifolia* is entomophilous and floral visitor’s dependant. *J. rhombifolia* is entomophylous, although spontaneous autogamy could favour reproduction in the absence of pollinators. *S. longifolia* could be an ambophilous species (pollinated both by insects and by the wind). This dual system may be the result of a system flexibility mechanism or an evolutionary transition. (Torreta and Basilio 2013).

Another relict forest ecosystem in Buenos Aires is the locally named Monte Blanco. This forest, that originally occupied the elevated border areas of the Low Delta islands of the Río Paraná and some areas of Río de La Plata river coast, has almost been eliminated as a consequence of unsustainable timber harvesting carried out in the region during the past century. At present, only relict patches with scarce regional representation may be found along the north-eastern part of the Río de La Plata shore. Many of these forests have been abandoned, resulting in secondary forest formation that is subject to numerous invasive exotic species, such as *Ligustrum lucidum*, *Morus alba*, *Rubus ulmifolius*, *Gleditsia triacanthos*, *Fraxinus pennsylvanica* and other species. A priori observations suggest that successional trends do not lead to recovery of the original forest. Because of the low density of native tree species, it is difficult to predict their future persistence. In conclusion, ecological restoration strategies will be needed in order to increase native tree species richness and forest biodiversity in the Lower Delta of the Paraná River, the original forest, locally referred to as the Monte Blanco forest (Kalesnik et al. 2008).



Figure 1. Location of natural forests (Talares and Monte Blanco) in Buenos Aires Province, Argentina.

Methods for conservation and propagation of native forest species from Talares and Monte Blanco

At the Facultad de Ciencias Agrarias y Forestales (FCAyF), Universidad Nacional de La Plata (UNLP), we carried out an investigation about the propagation requirements of indigenous tree species from Buenos Aires Talares and Monte Blanco forests. We adjusted the system of propagation by seeds, cuttings and/or *in vitro* morphogenesis in some of these trees species:

- *Celtis ehrenbergiana* Gill. ex Planch “tala”, Ramilo and Abedini (2007) achieved rooting of cuttings by using 50 ppm NAA. After 5 months, higher survival (71%) was recorded for NAA treatment (50 ppm) versus a mean survival of 2,5% when IBA was used (Figure 1).

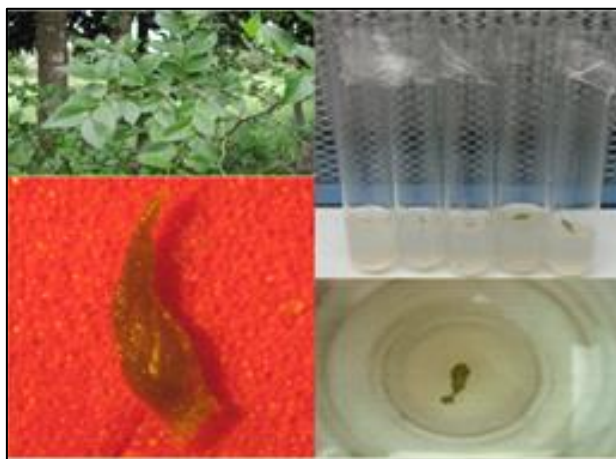


Figure 1. Micropropagation *Celtis ehrenbergiana*

- *Salix humboldtiana* “South American Native Willow” We improved the macro and micropropagation of this species (Adema et al. 2010; 2014). Macro propagation was achieved by rooting of cuttings (30 cm. long and 0.8 - 1.5 cm in diameter), immersed in IBA during 24 hours. *In vitro* rooting occurs on WPM supplemented with 0.1 mg. L⁻¹ of IBA (Figure 2).



Figure 2. Macropropagation of *Salix humboldtiana*

- *Parkinsonia aculeata* “Cina-cina”, micropropagation was achieved from nodal segments in BTM medium (Chalupa 1983) with KIN. The macropropagation can be obtained from juvenile cuttings collected during springtime.
- *Erythrina crista-galli* L. “Ceibo”, the *in vitro* protocol for Ceibo regeneration was adjusted from nodal sections of seedlings, which were obtained from seeds, germinated *in vitro*. The shoots formed roots in WPM medium with 0.1 mg. L⁻¹ of NAA. (Ruscitti and Abedini 1996).

- *Terminalia australis* “Palo Amarillo”, macropropagation was achieved by using cuttings (200 mm long and 8 mm in diameter on average) immersed in NAA solutions (Aquila et al 2001).
- *Acacia caven Molina* “Espinillo”, a micropropagation system was developed using somatic embryogenesis. The explants were cotyledons obtained from mature seeds (Marinucci et al. 2003; 2005).
- *Phytolacca tetramera* “Ombusillo”, micropropagation was possible via organogenesis from nodal and internodal sections cultivated on MS culture medium, supplemented with sucrose and IBA and BAP (Basiglio Cordal and Sharry 2012; Basiglio Cordal et al. 2014).
- *Phyllanthus sellowianus* “Sarandí blanco” Rivas and Abedini (1996) improved the micropropagation through direct organogenesis induced from nodal and internodal segments.
- *Citharexylum montevidense* “Espina de bañado”, macropropagation was possible by using cuttings from 7 mm. in diameter and with material less than 1 year old after 110 days (Roussy et al. 2011) (Figure 3).



Figure 3. Cuttings of *Citharexylum montevidense*

- *Scutia buxifolia* “coronillo” stem cuttings collected in winter were submerged for a 24 hr period in 5 concentrations of IBA and 5 of NAA resulting in 47 % survival with IBA (Ramilo and Abedini, unpublished).

We describe evidence of protocols through the major methods being used, developed and applied to propagate and conserve native species.

Conclusion

We describe challenges associated with the restoration of threatened trees in the Talaes and Monte Blanco forests of Northeast of Buenos Aires and analyse the effectiveness of methods used to define target species. We identified seed sources and generated information on the biology of rare or threatened tree species. Despite the many challenges associated with collection of seeds and to adjust propagation systems from rare and threatened native species, our data represent a significant first step towards improving the genetic diversity and species richness of the seedlings produced in the Buenos Aires forests. The methods described here may be of relevance to ecological restoration programmes elsewhere where the challenges of including rare and threatened tree species in a germplasm bank, seedling production and planting operations are likely to be similar.

References

- Adema M., Basiglio Cordal M., Briones V., Villareal B., Ciocchini G., Abedini W., Sharry S. (2014). Macro and micropropagation of *Salix humboldtiana* and *Salix babilónica*. Evaluation of the removal capacity of Cu. Jornadas de Salicaceae 2014- IV International Congress of Salicaceae, Argentina.
- Adema M., Curuchet G., Abedini W., Sharry S., Briones V., Villareal B., Basiglio Cordal MA.,

- Ciocchini G. (2010). Preliminary study of copper phytoremediation using *Salix humboldtiana*, Acts VII Latin American and Caribbean Encounter on Biotechnology.
- Aquila S. V., Bonicelli V., Carparo S., Rossi J., Castro M. T., Spegazzini E., Abedini W. (2001) Vegetative Propagation of *Terminalia Australis* “Palo Amarillo,” *Combretaceae*, Proceedings IV International Congress of Medicinal Plants.
- Arturi M (1997) Regeneration of *Celtis tala* Gill ex Planch in the NW of the Province of Buenos Aires. (Ph.D. thesis) School of Agriculture and Forestry Sciences, National University of La Plata, Buenos Aires. SEDICI Web <http://sedici.unlp.edu.ar/handle/10915/4658>.
- Arturi M. F., Goya J. F. (2004) Structure, dynamics and management of the talares of the NE of Buenos Aires. In: Arturi M., Frangi J., Goya J. (eds.), Ecology and Management of Argentina's forests EDULP, Argentina, 10:1-23
- Basiglio Cordal M., Sharry S. (2012) Conservation of medicinal species from the Province of Buenos Aires, the case of Ombusillo. *Bioethics Notebooks*. Ed. Ad Hoc, Argentina 17: 45 – 47.
- Basiglio Cordal M., Adema M., Briones V., Villarreal B., Panarisi M., Abedini W., Sharry S. (2014) Induction of somatic embryogenesis in *Phytolacca tetramera*, medicinal species of Argentina. *Emirates J Food Agric* 26(6): 552 - 557.
- Cabello M., Arambarri A. (2002) Diversity in soil fungi from undisturbed and disturbed *Celtis tala* and *Scutia buxifolia* forests in the eastern Buenos Aires province (Argentina). *Microbiol Res* 157:115–125.
- Cabrera A (1976) Argentine phytogeographic regions. *Encyclopedia of agriculture and gardening Argentina*. ACME S.A.I.C. Buenos Aires.
- Chalupa V. (1983) Micropropagation of Conifer and Broadleaved Forest Trees. *Communicationes Instituti Forestalis. Cechosloveniae* 13: 7-39.
- Goya J., Placi L., Arturi M., Brown A. (1992) Distribution and structural characteristics of the talares of the Biosphere Reserve “Parque Costero del Sur”. *Agronomics Faculty Journal*. La Plata 68: 53-64.
- Kalesnik F., Valles L., Quintana R., Aceñolaza P. (2008) Relictual Patches of Monte Blanco Forest en Delta del Río Paraná region *Serie Misc. INSUGEO-CONICET* 17:169 – 193.
- Parodi L. R. (1940) Native forests of Buenos Aires province. *Proc Natl Sci Acad* 7: 97-90.
- Ramilo D.I., Abedini W.I. (2007). Vegetative propagation of *Celtis tala* Gill. Ex Planch by rooting wooden stakes semilignified. XXI Jornadas Forestales de Entre Ríos. Argentina.
- Ramilo D.I., Abedini W.I (unpublished). Vegetative propagation of *Scutia buxifolia* Reissek by rooting wooden stakes semilignified, internal report. CEPROVE. School of Agriculture and Forestry Sciences.
- Ribichich A., Protomastro J. (1998) Woody vegetation structure of xeric forest stands under different edaphic site conditions and disturbance histories in the Biosphere Reserve Parque Costero del Sur, Argentina *Plant Ecol* 139 (2):189–201.
- Rivas M.C., Abedini W. (1996) Rapid clonal propagation of *Phyllanthus sellowianus* (Sarandí blanco). Third International Symposium on in vitro culture and Horticultural Breeding. Jerusalem, Israel.
- Roussy L., Pincirolí L., Briones V., Sharry S., Ciocchini G., Abedini W. (2011) Rooting of cuttings of different diameter of *Citharexylum montevidense* (Spreng) Moldenke, forest species native to the flora of Buenos Aires. V Latinamerican Forestry Congress, Lima, Peru. CONFLTA. La Molina University.
- Ruscitti M, Abedini W (1996) In vitro cloning of *Erythrina crista-galli* ecotypes resistant to environmental contamination. XVI Congress of Phytogenetics. Mexican Society of Phytogenetics (SOMEFI). Institute of Genetic Resources and Productivity of the Postgraduated School of Montecillo, Mexico.
- Torretta J., Basilio A. (2013) Pollen dispersion and reproductive success of four tree species of a xerophytic forest from Argentina. *Rev de Biología*. <http://dx.doi.org/10.15517/rbt.v57i1-2.11321>.