



# Improvement of isotope-based climate reconstructions in Patagonia through a better understanding of climate influences on isotopic fractionation in tree rings

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## ABSTRACT

Very few studies of stable isotopes exist across the Andes in South America. This study is the first presenting annually resolved chronologies of both  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  in *Nothofagus pumilio* and *Fitzroya cupressoides* trees from Northern Patagonia. Interannual variability in  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  was assessed over the period 1952–2011. Based on these chronologies, we determined the primary climatic controls on stable isotopes and tree physiological responses to changes in atmospheric  $\text{CO}_2$  concentrations ( $c_a$ ), temperature and humidity. Changes in specific intrinsic water use efficiency (iWUE) were inferred from variations in  $\delta^{13}\text{C}$  whereas the effects of  $\text{CO}_2$  increase on stomatal conductance were explored using  $\delta^{18}\text{O}$ . Over the 60-year period, iWUE increased significantly (by ca. 25%) in coincidence with the rise of  $c_a$ . The two species appear to have different strategies of gas-exchange. Whereas iWUE variations were likely driven by both stomatal conductance and photosynthetic assimilation rates in *F. cupressoides*, they were largely related to stomatal conductance in *N. pumilio*. After removing the low-frequency trends related to increasing  $c_a$ , significant relationships between  $\delta^{13}\text{C}$  and summer temperatures were recorded for both species. However,  $\delta^{13}\text{C}$  variations in *F. cupressoides* were more strongly influenced by summer temperatures than in *N. pumilio*. Our results advocate for an indirect effect of summer temperatures on stable isotope ratios, which is mostly influenced by sunlight radiation in *F. cupressoides* and relative humidity/soil moisture in *N. pumilio*.  $\delta^{13}\text{C}$  variations in *F. cupressoides* were spatially correlated to a large area south of 35°S in southern South America. These promising results encourage the use of  $\delta^{13}\text{C}$  variations in *F. cupressoides* for reconstructing past variations in temperature and large-scale circulation indexes such as the Southern Annular Mode (SAM) in the Southern Hemisphere.

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## 1. Introduction

Several attempts to document past climate variability in subtropical and temperate regions of South America have been conducted over the last decades. Long-term, climate sensitive dendrochronological records from the outer tropics to southern South America (e.g. Boninsegna et al., 2009) provide unique opportunities to reconstruct climate and large-scale atmospheric circulation patterns (e.g. Villalba et al., 2012; Morales et al., 2015). The use of tree-ring width (TRW) variations as tracers of past climate

fluctuations has significantly expanded our understanding of climate variability over the South American continent during the past centuries. However, instabilities in the relationships between climate and TRW have been identified in some records, with changes mostly coincident with major shifts in climate regime (Álvarez et al., 2015; Lavergne et al., 2015; Suarez et al., 2015). These decouplings have been observed in regions strongly influenced by both the Southern Annular Mode (SAM) and the El Niño Southern Oscillation (ENSO) variability patterns (Villalba, 2007), which questions the use of TRW for reconstructing long-term climatic variations across southern South America.

A recent study has highlighted the potential of the isotopic composition of oxygen in cellulose extracted from *Nothofagus pumilio* tree-rings to reflect variations in December–May temperatures ( $T_{\text{Dec-May}}$ ) over a large region in the Patagonian Andes

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south of 38°S ( $r = 0.57$ ,  $p < 0.05$ ; Lavergne et al., 2016). It has also shown that the relationship between  $\delta^{18}\text{O}$  in *Fitzroya cupressoides* and  $T_{\text{Dec-May}}$ , thought significant, was weaker than in *N. pumilio* ( $r = 0.30$ ,  $p < 0.05$ ). Due to the poor performance of  $\delta^{18}\text{O}$  in *F. cupressoides* as a climate proxy, the use of  $\delta^{18}\text{O}$  does not seem to increase the dendrochronological potential of this multi-millennium species with individuals exceeding 3600 years in age (Lara and Villalba, 1993). However, previous studies have shown that temperature and precipitation variations have a strong imprint on the carbon isotopic composition ( $\delta^{13}\text{C}$ ) of the tree-rings in both *N. pumilio* and *F. cupressoides* species (Leavitt and Lara, 1994; Srur et al., 2008; Urrutia-Jalabert et al., 2015).  $\delta^{13}\text{C}$  in tree-rings depend on factors affecting the photosynthetic uptake of  $\text{CO}_2$  and are mainly controlled by stomatal conductance ( $g$ ) and the rate of carboxylation during photosynthesis ( $A$ ) (Farquhar et al., 1989). These factors influencing isotopic fractionation are closely related to meteorological conditions suggesting that  $\delta^{13}\text{C}$  in *F. cupressoides* and *N. pumilio* could be considered targeted climate proxies in Patagonia. These studies have also shown that the climatic information encoded in  $\delta^{13}\text{C}$  ratio is specific and, in some case, site dependent. Consequently, an improved understanding of the specific physiological responses to local and regional environmental conditions is, therefore, a prerequisite for an accurate interpretation of  $\delta^{13}\text{C}$  in tree rings.

Variations in the intrinsic water-use efficiency (i.e. the ratio of carbon gain to water loss;  $i\text{WUE} = A/g$ ) calculated using tree-ring  $\delta^{13}\text{C}$  provide insights on tree's physiological responses to climate (e.g. Frank et al., 2015; Wieser et al., 2016). This isotopic approach has been widely used in several regions, showing a consistent enhancement in  $i\text{WUE}$  over the 20th century at most sites (e.g. Lévesque et al., 2014; Saurer et al., 2014). However the specific nature of changes in  $i\text{WUE}$  (due to variations in water availability, in the efficiency of  $A$  or in  $g$ ) cannot be precisely determined using  $\delta^{13}\text{C}$  alone (e.g. Scheidegger et al., 2000). Since  $\delta^{18}\text{O}$  is not modulated by  $A$ , the information on evaporative enrichment recorded in  $\delta^{18}\text{O}$  can be useful to discriminate the effects of  $A$  and  $g$  on  $i\text{WUE}$  (Scheidegger et al., 2000; Roden and Siegwolf, 2012). Therefore, the joint analysis of  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  in tree-rings can be used to distinguish the effects of  $g$  and  $A$  on  $i\text{WUE}$  variations, and therefore, to disentangle the different gas-exchange strategies of tree species under changing ambient  $\text{CO}_2$  and environmental conditions (e.g. Nock et al., 2011).

The main goals of our study were twofold: 1) to characterize the physiological responses of *Fitzroya cupressoides* and *Nothofagus pumilio* to environmental changes for improving our interpretation of  $\delta^{13}\text{C}$  in tree rings, and 2) to evaluate the performance of the carbon isotopic composition of the cellulose in these two species as a paleoclimate proxy. For reaching these goals, we developed five annually resolved  $\delta^{13}\text{C}$  chronologies covering the last 60 years (1952–2011) from sites at different elevations in Northern Patagonia (41°S), and combined them with  $\delta^{18}\text{O}$  chronologies from Lavergne et al. (2016). The main novelty of our approach lies in the simultaneous examination of both isotopes of two species in southern South America. Only by improving our knowledge about the forcings of the isotopic variability in tree-rings and the eco-physiological information recorded therein, we will be finally able to improve stable-isotope based climate reconstructions from tree-rings.

## 2. Materials and methods

### 2.1. Regional setting and climate

The regional climate along the Patagonian Andes is mainly driven by the interactions between the circum-Antarctic cyclonic belt to the south and the subtropical Pacific high-pressure cell

to the northwest. The strong Westerlies coming across the Pacific Ocean permanently interact with the north–south mountain range of the Andes (e.g. Garreaud et al., 2013). The air masses from the Pacific lose moisture during the ascent on the western slope of the Andes and descend drier on the eastern foothills. Therefore, the mountain range induces a dramatic decline in precipitation from total annuals of about 4000 mm near the continental divide to less than 500 mm toward the Patagonian forest-steppe ecotone (Villalba et al., 2003). At Bariloche, in the eastern side of the gradient (41°12'S–71°12'W, 840 m asl), the mean annual temperature is 8.5 °C, with relatively cold winters (mean JJA temperature ~3 °C) and mild summers (mean DJF temperature ~14.2 °C). Seasonal variations in the Pacific anticyclone positions induce strong precipitation seasonality. Precipitation is largely concentrated from late fall to early spring followed by a drier period during summer and early fall (López Bernal et al., 2012).

### 2.2. Sampling and tree-ring processing

Sampling was conducted in March 2013 in five sites in northern Patagonia. Two sites for *F. cupressoides* (950–1050 m elevation) and three sites for *N. pumilio* (1270–1610 m) were selected in Parque Nacional Nahuel Huapi, near Bariloche (at 41°05'S–71°21'W, see Lavergne et al., 2016 for details). Cores were taken at breast height (1.3 m above the ground) using a 5 mm diameter Pressler increment borer. The core samples were dated to the calendar year of their formation following the techniques described by Stokes and Smiley (1968) and ring widths were measured at an accuracy of 0.001 mm. Cross-dating of growth rings to detect any error in the measurements was based on more than 60 cores from 30 or more trees at each site. Quality-control and accuracy of measurements and cross-dating were conducted with the COFECHA program, which allows the matching of ring-width among radii within a tree and among different trees (Holmes, 1983).

According to the procedure described in Lavergne et al. (2016), 6 trees for *F. cupressoides* and 4 trees for *N. pumilio* were selected for building the isotopic chronologies at each sampling site. All trees from each site were pooled to produce an average  $\delta^{13}\text{C}$  series at each site. Annual tree rings were split using a scalpel and pooled year by year. The wood samples were then chipped and grounded in a ball mill for homogenization.  $\alpha$ -cellulose was extracted from the wood according to the SOXHLET chemical method derived from Leavitt and Danzer (1993).  $\alpha$ -cellulose was homogenized ultrasonically with a sonotrode apparatus and freeze-dried. Cellulose samples of around 0.10 mg were loaded in tin-foil capsules. The  $\delta^{13}\text{C}$  was determined with an elemental analyser (EA NC2500, Carlo Erba) coupled with a Finnigan MAT252 mass spectrometer. An internal laboratory reference of cellulose (Whatmann® CC31) was used to correct for instrument drift and to normalize the data to internationally accepted standards. Along the sequence analyses, the isotopic composition of CC31 was measured every three samples. The standard deviation (SD) obtained from the measurement of the isotopic composition of 10 consecutive CC31 standards was typically  $\pm 0.1\%$ . The analytical procedure for  $\delta^{18}\text{O}$  measurements is described in Lavergne et al. (2016). The analyses of both stable isotopes on each sample were repeated at least once and up to three times. This methodology allows rejecting outlier measurements, which do not fit with the maximum accepted range.

### 2.3. Correction of non-climatic trends

The measured  $\delta^{13}\text{C}$  chronologies ( $\delta^{13}\text{C}_{\text{raw}}$ ) show a decreasing trend due to the rise of  $^{13}\text{C}$ -depleted atmospheric  $\text{CO}_2$  induced by fossil fuel burning and deforestation since industrialization (the Suess effect; Keeling, 1979). Therefore, the  $\delta^{13}\text{C}_{\text{raw}}$  series were corrected for the anthropogenic influence by removing the preindus-

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