# Is there a relationship between the concentration of nutrients in leaves and the class of site where *Nothofagus antarctica* forests growth in southern Patagonia, Argentina?

¿Existe relación entre la concentración de nutrientes en hojas con la clase sitio donde se desarrollan los bosques de *Nothofagus antarctica* en Patagonia sur, Argentina?

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#### **SUMMARY**

The objective of this study was to determine whether differences in site quality correlate with better leaf nutritional status in pure *Nothofagus antarctica* (ñire) forests. Nine pure ñire stands of the three site classes (SC) were selected to represent the most representative sites (97 % of the total area of these forests) of the southern Patagonia region. At each study site, fully expanded leaves (mature and green) were collected from five dominant trees in each stand. These leaves were washed and subsequently oven dried, weighed, and ground prior to macro (C, Ca, K, Mg, N, P, and S) and micro element (Al, Fe, Mn, and Na) determination. Additionally, three composite samples (from five subsamples) of the first 20 cm of soil were collected from each site and processed for macro- and micro-element determination. Macro-nutrient concentrations in leaves varied between site classes (SC) in some elements such as K, Mg, N, P, and S, with a trend to be lower in the worse SC. In the case of soil macronutrients, all the evaluated elements showed the lowest values in the SC 5 stands. Thus, soil and leaf concentrations were significantly and positively correlated with N, P, and S. Conversely, Al, Fe, and Mn concentrations in leaves were lower in the best site class stand. With this information, we can suggest that the best classes of forests in southern Patagonia were positively associated with higher values of some macronutrients and negatively associated with some microelements.

Keywords: forest, soil, ñire, dominant height, leaves.

## RESUMEN

El objetivo de este estudio fue determinar si la diferencia en la calidad del sitio se correlaciona con un mejor estado nutricional foliar en bosques puros de *Nothofagus antarctica*. Se seleccionaron nueve rodales puros de *N. antarctica* de tres clases de sitio (CS), expresando los sitios más representativos (97 % de la superficie total de estos bosques) de la región sur de la Patagonia. En cada sitio de estudio se recolectaron hojas completamente expandidas (maduras y verdes) de cinco árboles dominantes en cada rodal. Estas hojas fueron lavadas, posteriormente secadas al horno, pesadas y molidas antes de la determinación de macro (C, Ca, K, Mg, N, P y S) y microelementos (Al, Fe, Mn y Na). Además, se recogieron tres muestras compuestas (de cinco submuestras) de los primeros 20 cm de los suelos de cada lugar y se procesaron para la determinación de macro y microelementos. Las concentraciones de macronutrientes en hojas variaron entre CS en algunos elementos como K, Mg, N, P y S, con una tendencia a ser menores en las peores CS. En el caso de los macronutrientes del suelo, todos los elementos evaluados mostraron los valores más bajos en los rodales de peor CS. Así, las concentraciones en suelo y hojas estuvieron significativa y positivamente correlacionadas para N, P y S. Por otro lado, las concentraciones de Al, Fe y Mn en hojas fueron menores en el rodal de mejor clase de sitio. Con esta información podemos sugerir que las mejores clases de bosques de ñire en el sur de la Patagonia están asociadas positivamente con valores más altos de algunos macro nutrientes y negativamente con algunos micro elementos.

Palabras clave: bosque, suelo, ñire, altura dominante, hojas.

## INTRODUCTION

In general, net primary production in temperate forest ecosystems is mainly driven by climatic factors and internal nutrient cycling, being the litter fall the main pathway for nutrient input into soil. Low nutrient requirements had been raised as an important plant strategy to conserve nutrients in this type of ecosystems (Aerts and Chapin 1999).

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In the south hemisphere, the Nothofagus (Fagaceae) is one of the most representative tree genus of the temperature forests, where N. antarctica (G. Forster) Oerst. (ñire, ñirre) is the most broadly distributed species from 36° 30' to 56° 00' of S latitude and from 0 to 2,000 m a.s.l., (Donoso et al. 2006). This implies that nire grows in a very diverse range of environmental conditions in Chile and Argentina. In continental southern Patagonia (Santa Cruz province, Argentina), ñire covers 160,000 ha, growing in contrasting environmental conditions such as poorly drained sites with high precipitations in the Andean mountains or drier sites at lower elevations and exposed to strong winds in the limit with the Patagonian steppe (Peri and Ormaechea 2013). This range of environmental conditions consequently has been associated with different net primary productivity of these forests. Related to this, Ivancich et al. (2011) have characterized the N. antarctica forests productivity through site quality index equations and proposed a "site class" (SC) classification using the height of dominant mature trees as a proxy of nire forest productivity. According to this, in the best SC stands, dominant tree height reaches up to 14 m (SC 1) and in lowest SC dominant tree height is less than 8 m (SC 5). Furthermore, several studies related SC with different ecological and productive features such as carbon accumulation (Peri et al. 2010), understory productivity, litter-fall and response to water shortage (Bahamonde et al. 2015, 2019a). Also, low rates of litter decomposition and soil nitrogen mineralization have been reported for these forests, although these studies have been carried out only in the lowest site classes (Bahamonde et al. 2013). On the other hand, Bahamonde et al. (2018) have evaluated several soil and climatic features and their relative importance on SC of N. antarctica forests in southern Patagonia. They reported that better SC (higher dominant trees) were related to locations with low altitudes, less differences between maximal and minimal temperatures, and sandy soils. Regarding the variation in nutrient concentration in ñire stands growing in different site classes, information is very scarce. Peri et al. (2006, 2008) have reported that nutrient accumulation and concentration in ñire leaves was low in a stand growing in a marginal site class (dominant mature tree height of 5.2 m). However, these studies only focused in variations within stands.

Frangi et al. (2005) studied the nutrient cycling in Nothofagus pumilio (closely related to N. antarctica) forests growing at different altitudes in Tierra del Fuego, which included the highest elevations for these forests (timberline) broadly known as krummholz. They reported that N. pumilio forests growing at krummholz showed higher nutrient concentrations and faster rates of nutrient fluxes compared to forests located at lower elevations. Also, N and P concentrations in mature leaves have been positively correlated with soil concentrations in several Nothofagus (including N. antarctica) of the Patagonian forests (Diehl et al. 2008). Hevia et al. (1999) reported differences in leaf N and P concentrations between three Nothofagus

sp. (*N. pumilio, N. dombeyi* and *N. obliqua*) in Chile, but not clear association with the elevation were found.

On the other hand, other chemical elements that are not considered nutrients are of interest to be studied in *Notho-fagus* forests, such as aluminium (Al) and sodium (Na). In the case of Al, it is known that there are concentration levels that could be toxic to plants (Kabata-Pendias 2011, Kirkby 2012). While Na has been described as a beneficial element for plants (Pilon-Smits *et al.* 2009). The presence of both elements has already been reported in leaves of grasses and *Nothofagus* in Patagonia (Bahamonde *et al.* 2016, 2019b).

However, little is known about the incidence of soil and climatic features that determines different site classes on the nutritional status of *N. antarctica* forests in southern Patagonia. In this context, the objective of this study is to answer the following question: Does the difference in site quality correlate with higher nutrient concentrations in pure *N. antarctica* forests? We hypothesized that stands growing in worse site classes will have lower nutrients and micro elements concentrations in leaves compared to the better sites.

#### **METHODS**

Study sites. An exploratory survey was performed analysing the map of site class distribution of N. antarctica forests in Santa Cruz province (southern Patagonia) (Peri and Ormaechea 2013) and including sites which are part of PEBANPA network (Parcelas de Ecología y Biodiversidad de Ambientes Naturales en Patagonia Austral) (Peri et al. 2016). Thus, we selected pure N. antarctica stands of the three site class (Ivancich et al. 2011) representing the most representative sites (97 % of the total area of these forests) of the region (SC 3, 4 and 5) (Peri and Ormaechea 2013). According to the classification proposed by Ivancich et al. (2011) in SC 3 height of mature dominant trees are in the range 10 - 12 m; SC 4: 8 - 10 m; and SC 5 < 8 m. Three mature stands of each SC were selected for sampling. Main climatic, productive and location features of each evaluated stand are given in table 1. Climatic parameters for each site were derived from the WorldClim data set (Hijmans et al. 2005, WorldClim 2005), meanwhile the altitude was obtained from Farr et al. (2007).

Sampling and measurements. During March 2015, at each study site, we measured in a 500 m<sup>2</sup> plot the height of three mature dominant trees to determine the site class according to Ivancich *et al.* (2011) classification as an indicator of productivity. At the same time, fully expanded leaves (mature and green) were collected from five dominant trees at each stand. In each sampled tree the leaves were taken from the four cardinal points and considering that leaves were from fully sunny exposed branches/leaves at height 2.0–2.5 m above soil surface. The leaf samples were immediately taken to the lab in closed plastic bags and kept at

3 °C in the refrigerator until processing. Leaf tissues were carefully washed with 0.1 % detergent (Cif) to remove surface contaminants and then rinsed with abundant distilled water. These were subsequently oven dried at 70 °C for 2 days, weighed and grounded prior to macro (C, Ca, K, Mg, N, P and S) and micro elements (Al, Fe, Mn and Na) determination. Nitrogen was measured with an elemental analyzer (TruSpec). The remaining elements were determined by inductively coupled plasma (ICP) analysis (Optima 3000, PerkinElmer) following the UNE-EN ISO/IEC 17,025 standards for calibration and testing laboratories (CEBAS-CSIC Analysis Service, Murcia, Spain). Additionally, three composite samples (from five subsamples) of the upper 20 cm of soils were collected from each site, in places where leaves were also sampled. Samples were air dried, screened through 2 mesh, oven dried at 40 °C and sieved through 20 mesh. Soil pH was determined in saturated soil paste (30 ml deionized water per 100 g soil). Carbon and nitrogen concentrations of soil samples were determined by dry combustion using a LECO CNS-2000 (Tabatabai and Bremner 1991). The remaining elements were determined by inductively coupled plasma (ICP) analysis as above described for leaves samples.

Statistical analysis. A multiple linear regression analysis was conducted to evaluate the relationship between dominant mature trees height (DH) (dependent variable) and four independent variables: altitude, mean annual precipitation (MAP), mean annual temperature (MAT), and soil pH. The objective was to determine how these environmental factors influence DH. The model was assessed using the coefficient of determination (R<sup>2</sup>), the adjusted R<sup>2</sup>, and the F-statistic to determine its overall significance. Individual predictor significance was evaluated using t-tests and p-values. Additionally, residual diagnostics were performed, including:

- Normality test using the Shapiro-Wilk test on residuals
- Multicollinearity assessment through the Variance Inflation Factor (VIF).
- Residual plots to check for homoscedasticity and linearity.

Exploratory testings were carried out to verify the compliance with the assumptions of normality, homoscedasticity and independence of data for each evaluated situation. While the Shapiro–Wilk test was performed to verify the normality of the data, the Levene test was used to verify homoscedasticity. The independence was verified by analysing residuals from graphs. Leaves and soil nutrient concentrations were analysed with one way ANOVA with site classes as main factor. Tukey tests were carried out to test differences between factors when F-values were significant (P < 0.05). Pearson correlation analyses were carried out to evaluate the association

between soil and leaves elements and between soil pH and soil elements. Also, a principal component analysis (PCA) was performed to analyse the influence of the mineral concentrations (soil and leaves) on Site Class over the sample distribution in an ordination space. PCA included Monte Carlo permutation test (n = 999) to assess the significance of each axis.

Statistical analyses were performed using R (version 4.2.1, R Foundation for Statistical Computing, Vienna, Austria).

#### **RESULTS**

In general, there was a trend where dominant mature trees reached higher heights (better site class) at lower elevations. However, a multiple regression analysis with altitude, mean annual precipitation (MAP), mean annual temperature (MAT) and soil pH as independent variables and dominant tree height (DH) as the dependent variable showed that soil pH was the only significant variable that was positively correlated with DH (tables 1 and 2).

Macro nutrients concentrations in leaves varied between site classes (SC) in some elements such as K, Mg, N, P and S, with a trend to be lower in the worse SC (figure 1). Carbon and Ca concentrations did not vary between SC. On the other hand, C:N ratio was higher in SC 5 and N:P ratio was lower in SC 3. In the case of soil macro nutrients, all the evaluated elements showed lowest values in the SC 5 stands. Thus, soil and leaves concentrations were significantly and positively correlated for N, P and S (table 2).

On the other hand, Al, Fe and Mn concentrations in leaves were lower in the best site class stand (SC 3) compared with other stands growing in SC 4 and 5, but Na concentration was higher. The opposite was observed in soils, where the lowest concentrations of micro elements were in lower site classes (figure 2). Thus, a negative correlation was found between leaves and soil concentrations of Al and Fe (table 3). On the other hand, soil pH was positively correlated with the concentration of soil nutrients (N, K, P, S and Fe) and other elements such as Al (table 4).

The first three principal components explain a total of 57.8% of the variation in the dataset (PC1: 30.9 %; PC2: 16.1 % and PC3: 10.8 %). This indicates a moderate level of structure in the data, with PC1 and PC2 capturing nearly half of the variability. While not dominant, these axes still offer meaningful insights into the underlying gradients of variation. The first principal component (PC1), explaining 30.9 % of the total variance, is strongly associated with variables related to soil chemical composition, particularly high negative relationship of Nitrogen (N), Potassium (K) and moderate contributions of Phosphorus (P) and Sulphur (S). Site Class 5 appears to cluster along the high end of PC1, indicating lower soil N, K, P and S levels. SC3 and SC4 are more distributed toward the centre or lower range of PC1, suggesting higher N, K, P and S in soils. The second component (PC2), accounting for 16.13 % of the

**Table 1.** Main characteristics of *Nothofagus antarctica* forests located in contrasting environmental conditions, in southern Patagonia. Principales características de los bosques de *Nothofagus antarctica* ubicados en condiciones ambientales contrastantes, en Patagonia sur.

| Site Class | Geographical coordinates      | Altitude (m a.s.l.) | MAP (mm yr¹) | MAT (°C) | DH (m) | Soil pH |
|------------|-------------------------------|---------------------|--------------|----------|--------|---------|
| SC 3       | 51°56′57.6" S - 71°41′41.6" W | 130                 | 336          | 6.2      | 10.8   | 5.7     |
|            | 51°50′53" S - 71°46′18" W     | 138                 | 330          | 6.2      | 11.3   | 5.7     |
|            | 51°01′30.1" S - 72°13′11.2" W | 203                 | 291          | 6.6      | 11.0   | 5.6     |
| SC 4       | 51°13′21.0" S - 72°15′34.0" W | 295                 | 331          | 5.9      | 8.1    | 5.7     |
|            | 51°17′50.7" S - 72°11′2.18" W | 414                 | 334          | 5.3      | 8.8    | 5.6     |
|            | 49°17′22.4" S - 72°53′34.9" W | 416                 | 808          | 7.5      | 9.0    | 5.5     |
| SC 5       | 51°55′55.4" S - 71°39′23.7" W | 130                 | 335          | 5.6      | 7.2    | 5.1     |
|            | 49°14′16.3" S - 72°54′12" W   | 450                 | 821          | 7.3      | 4.2    | 5.0     |
|            | 51°57′22.7" S - 71°31′40.7" W | 141                 | 330          | 5.4      | 6.5    | 5.1     |

Site Class (SC) is determined according to Ivancich et al. (2011) classification. MAP: mean annual precipitation; MAT: mean annual temperature; DH: dominant height of mature trees.

La clase de sitio (SC) se determina según la clasificación de Ivancich et al. (2011). MAP: precipitación media anual; MAT: temperatura media anual; DH: altura dominante de los árboles maduros.

 Table 2. Regression model coefficients for DH (table 1) prediction.

Coeficientes de modelo de regresión para predicción de altura de árboles dominantes (tabla 1).

| Variable  | Coefficient | Standard error | t value | p-value |
|-----------|-------------|----------------|---------|---------|
| Intercept | -28.21      | 8.98           | -3.142  | 0.035   |
| Altitude  | -0.0061     | 0.005          | -1.135  | 0.320   |
| MAP       | -0.0020     | 0.007          | -0.301  | 0.779   |
| MAT       | 0.9007      | 1.277          | 0.706   | 0.519   |
| pН        | 6.1697      | 2.115          | 2.916   | 0.043   |

**Table 3.** Pearson correlation coefficient (R) and p-value (P) between elements concentration in leaves of *Nothofagus antarctica* and soils where the trees growth, in three contrasting sites in southern Patagonia.

Coeficiente de correlación de Pearson (R) y valor p (P) entre la concentración de elementos en hojas de Nothofagus antarctica y los suelos donde crecen los árboles, en tres sitios contrastantes del sur de la Patagonia.

|   | С     | N        | Al      | Ca   | Fe      | K        | Mg    | Mn    | Na    | P       | S       |
|---|-------|----------|---------|------|---------|----------|-------|-------|-------|---------|---------|
| P | 0.169 | 0.001    | 0.012   | 0.99 | < 0.001 | 0.053    | 0.129 | 0.842 | 0.347 | 0.005   | < 0.001 |
| R | 0.24  | 0.48 (+) | 0.37(-) | 0    | 0.50(-) | 0.27 (+) | 0.23  | 0.03  | 0.14  | 0.41(+) | 0.55(+) |

Signs in parentheses indicate if the correlation is positive (+) or negative (-).

Los signos entre paréntesis indican si la correlación es positiva (+) o negativa (-).

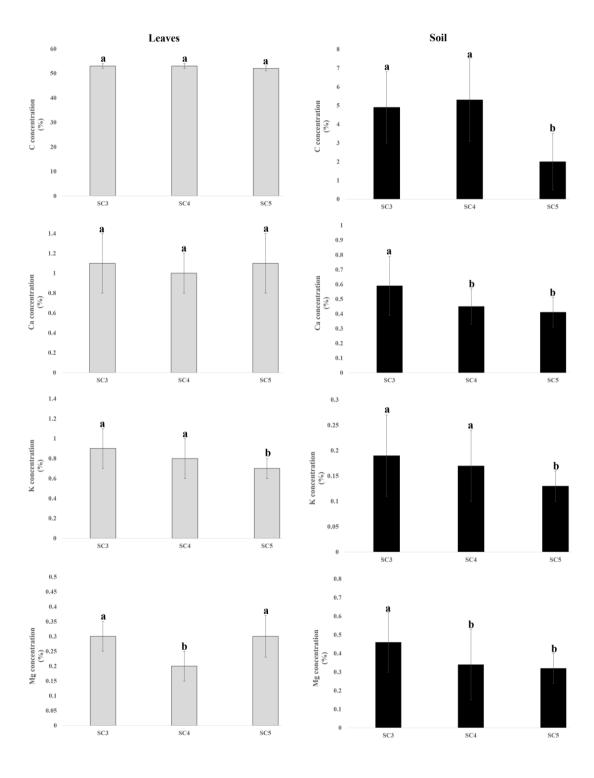
**Table 4.** Pearson correlation coefficient (R) and p-value (P) between soil pH elements concentration in soils where the *Nothofagus antarctica* trees growth, in three contrasting sites in southern Patagonia.

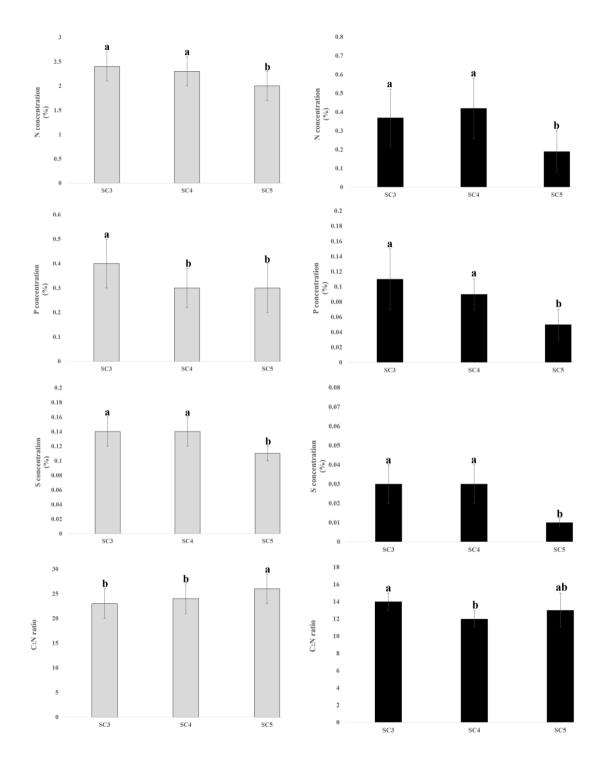
Coeficiente de correlación de Pearson (R) y valor p (P) entre pH y la concentración de elementos en los suelos donde crecen los árboles de *Nothofagus antarctica*, en tres sitios contrastantes del sur de la Patagonia.

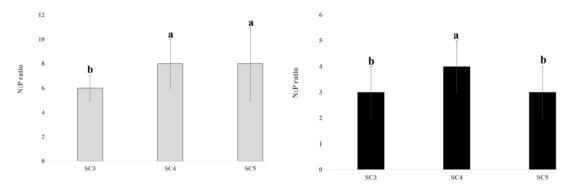
|   | С       | N        | Al      | Ca   | Fe      | K       | Mg    | Mn    | Na    | P       | S       |
|---|---------|----------|---------|------|---------|---------|-------|-------|-------|---------|---------|
| P | < 0.001 | < 0.001  | 0.015   | 0.75 | < 0.001 | 0.042   | 0.081 | 0.084 | 0.821 | < 0.001 | < 0.001 |
| R | 0.53(+) | 0.55 (+) | 0.53(+) | 0.38 | 0.46(+) | 0.46(+) | 0.33  | 0.44  | 0.04  | 0.67(+) | 0.54(+) |

Signs in parentheses indicate if the correlation is positive (+) or negative (-).

Los signos entre paréntesis indican si la correlación es positiva (+) o negativa (-).







**Figure 1.** Mean macro nutrients (C, Ca, K, Mg, N, P and S) concentrations and C:N, N:P ratios (± standard deviation) of *Nothofagus antarctica* mature leaves (left column, grey) and soils (right column, black) from three site classes (SC), in southern Patagonia. Site Class (SC) is determined according to Ivancich et al. (2011) classification. Different letters for a same element indicate significant differences between SC according to F (P) values.

Concentraciones medias de macro nutrientes (C, Ca, K, Mg, N, P y S) y relaciones C:N, N:P (± desviación estándar) de hojas maduras de *Nothofagus antarctica* (columna izquierda, gris) y suelos (columna derecha, negro) de tres clases de sitio (SC), en Patagonia sur. La clase de sitio (SC) se determina según la clasificación de Ivancich *et al.* (2011). Letras diferentes para un mismo elemento indican diferencias significativas entre SC según valores de F (*P*).

variance, is influenced primarily by foliar nutrient concentrations, particularly lower concentrations of Manganese (Mn) and Iron (Fe), and higher concentrations of Potassium (K) in leaves. SC3 samples tend to align with higher PC2 values. SC4 and SC5 are more variable along this axis, indicating site-specific differences in foliar nutrient dynamics.

#### DISCUSSION

In this study, a new contribution to the knowledge about the incidence of environmental factors (topography, soil and climate) on nutrient performance of N. antarctica forests is provided. This information may assist to a better understanding and practical management of these forests in a context of silvopastoral use, ecological significance and changing climate (Bahamonde et al. 2018). Despite some differences in leaves macro nutrients concentrations between SC, in general the values are in the range of previously published information for N. antarctica in southern Patagonia (Peri et al. 2006, 2008, 2010, Bahamonde et al. 2019b). The lower leaves concentrations of K, N, P and S in trees growing at SC 5 are clearly associated with low soil nutrient concentrations. The low leaves concentrations in a marginal site class, as SC 5, were also reported by Peri et al. (2008) for aerial components (branches, leaves, bark and wood) compared with macro nutrients concentrations in a better site class stand of ñire (Peri et al. 2006). However, Bahamonde et al. (2019b) did not detect differences in nutrient concentrations in ñire leaves from 3 stands with contrasting productivities (dominant tree heights). It should be noted that the study by Bahamonde et al. (2019b) also found no differences in most of the soil nutrients evaluated between sites. This study also discusses the complex interaction between soil characteristics and environmental variables that ultimately determine nutrient concentrations in leaves.

Concerning to the correlation between soil fertility and leaves concentrations, although in the studies of Peri et al. (2006, 2008) there were not differences in soil concentrations between site classes, there are antecedents in concordance with our results. For example, Diehl et al. (2003) found a positive significant correlation (R = 0.68) between soil and mature leaves N concentration in a study with 10 species (including N. antarctica) of native forest in northern Patagonia. Similarly, on another study Diehl et al. (2008) found positive correlations between soil and mature leaves N and P concentrations in native forests of Patagonia. Bahamonde et al. (2013) reported lower annual net soil nitrogen mineralization in a ñire forest growing in a SC 5 compared to stand growing in a better site class. Thus, the incidence of N and P availability in forest soils as limiting for tree growth is broadly recognized (Aerts and Chapin 1999). Moreover, N:P ratio in leaves is commonly used as an indicator, being the values proposed by Aerts and Chapin (1999) the mostly used in temperate forests, where N:P values less than 14 indicate N limitation; N:P values higher than 16 imply P limitation; and N:P values between 14 and 16 would indicate both N and P limitations. In the present study, all SC stands showed a clear limitation for N, coincident with Diehl et al. (2008) in Nothofagus forests (including N. antarctica) in northern Patagonia. Related to this, Frangi et al. (2015) reported a significant positive correlation between mean annual temperature and N:P ratio in an analysis which included all Argentinean forests. Bahamonde et al. (2019) reported N:P ratios under 14 for N. antarctica and N. pumilio forests growing in different environmental conditions in southern

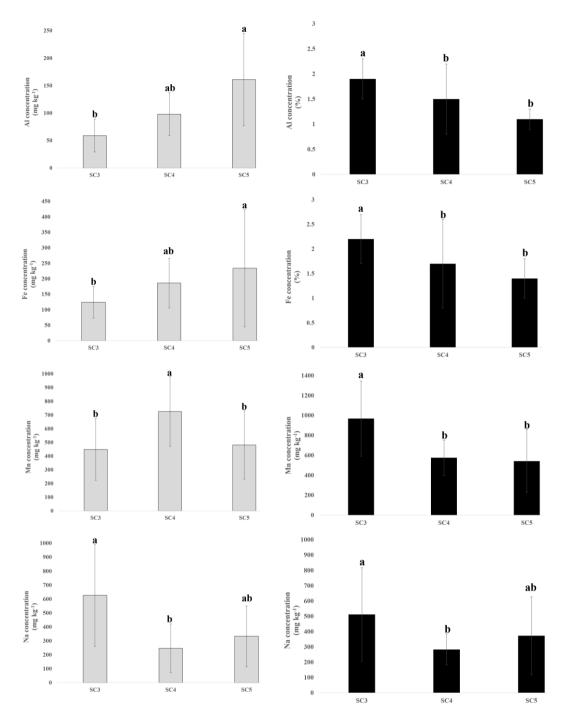


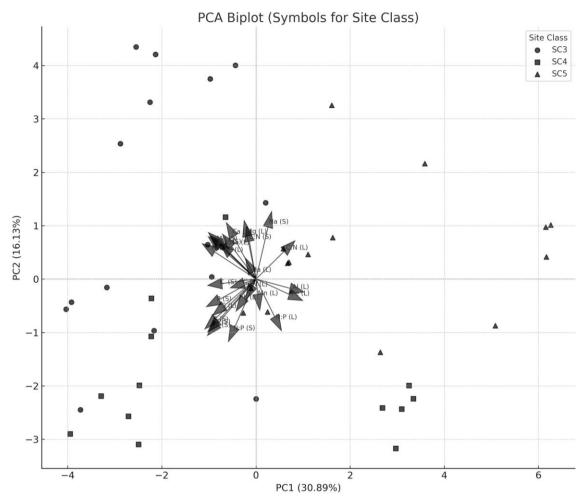
Figure 2. Mean micronutrients (Fe and Mn) and additional micro elements of interest concentrations (Al and Na) (± standard deviation) of *Nothofagus antarctica* mature leaves (left column, grey) and soils (right column, black) from three site classes (SC), in southern Patagonia.

Site Class (SC) is determined according to Ivancich et al. (2011) classification.

Different letters for a same element indicate significant differences between SC according to F (P) values.

Concentraciones medias de micronutrientes (Fe y Mn) y microelementos de interés adicionales (Al y Na) (± desviación estándar) de hojas maduras de *Nothofagus antarctica* (columna izquierda, gris) y suelos (columna derecha, negro) de tres clases de sitio (SC), en Patagonia sur. La clase de sitio (SC) se determina según la clasificación de Ivancich *et al.* (2011).

Letras diferentes para un mismo elemento indican diferencias significativas entre SC según valores de F (P).



**Figure 3.** Principal component analysis including leaves and soil variables related to site classes of *Nothofagus antarctica* forests in southern Patagonia. SC is the site class where each stand is growing.

Análisis de componentes principales, incluyendo variables foliares y edáficas, relacionadas con las clases de sitio de los bosques de *Nothofagus antarctica* en la Patagonia austral. SC representa la clase de sitio donde crece cada rodal.

Patagonia. These authors suggested that this N limitation in Patagonian forests was mainly due to very low organic matter decomposition and N mineralisation rates in these soil forests. In agreement with this, low decomposition of litterfall and roots, low N mineralisation and soil respiration have been reported in N. antarctica forests in southern Patagonia (Bahamonde et al. 2013, Peri et al. 2015, Bahamonde et al. 2016, Gargaglione et al. 2018). On the other hand, several mechanisms have been described in the literature to improve P acquisition in plants. The generation of cluster roots has been reported in Embothrium coccineum, a Proteaceae that grows in forests in Patagonia, as an efficient mechanism for P acquisition in situations of low availability (Lambers et al. 2012). Similarly, species such as Nothofagus have been documented to have established symbiotic relationships with mycorrhizal fungi that increase phosphatase activity and carboxylate exudation, which improve the availability and absorption of phosphorus in these plants (Gallardo et al. 2012). Related to this, Zhou

et al. (2020) have indicated that foliar Mn concentration as an indirect indicator of the use of carboxylates by roots to acquire phosphorus. In our study, the highest concentration of P in leaves (and lowest N:P ratio) occurred in SC 3, which had a lower concentration of Mn in leaves, which allows us to infer that its higher concentration of P would not be associated with the exudation of carboxylates, but probably with an increase in phosphatase activity due to its association with mycorrhizal fungi. Likewise, it has recently been published that mycorrhizal fungi in N. antarctica forests in Patagonia are more associated with soils with higher pH (Truong et al. 2024) as is the case of stands growing in SC 3 in our study.

Published data about micro nutrients concentrations in *N. antarctica* leaves are very scarce. Bahamonde *et al.* (2019) found Al, Fe and Mn concentrations in the range of the values of the present study in ñire leaves of *N. antarctica* and *N. pumilio* mixed forests in southern Patagonia. Also, Bahamonde *et al.* (2016) published micro elements

concentrations in grasses which were growing in N. antarctica soil forests in southern Patagonia. Compared to our results, these authors reported lower concentrations of Al, Fe, Mn and Na in grasses than N. antarctica leaves. Both Al and Fe leaves concentrations were higher in SC 5, despite the concentrations of these elements were lower in SC 5. Nevertheless, these results would be related to soil pH, with the lowest value in this site class (SC 5), supporting the well-known negative correlation between soil pH and Al and Fe availability for plants (Marschner and Römheld 1996). On the other hand, both Mn and Na in leaves did not show a clear trend related to neither SC or soil fertility. Regardless of differences between site classes, all the evaluated micro elements (Al, Fe, Mn and Na) were in the range of the concentration considered as sufficient for growing requirements by the literature (Kirkby 2012). Also, the concentrations of these elements were under the range of the considered toxic concentrations for the available studies in the literature (Kabata-Pendias 2011).

## CONCLUSIONS

Most of macro nutrients (K, N, P and S) show a trend of higher values in leaves of trees growing in better site classes, being positively correlated with soil nutrient concentrations and pH of soil. The most limiting macro nutrient in these forests is N, which is concordant with previous research in these forests. On the other hand, some micro elements, as Al (not considered as a nutrient) and Fe (nutrient), are higher in leaves of stands growing in worse site classes and negatively correlated with their soil concentrations. With this information we can suggest that the best classes of ñire forests in southern Patagonia are positively associated with higher values of some macro nutrients and negatively associated with some micro elements.

# **AUTHORS CONTRIBUTIONS**

Héctor Bahamonde participated in the generation of the idea, did much of the field sampling, data analysis and writing of the article. Jorge Frangi contributed with the experimental design and reviewed the article. Pablo Peri was part in the generation of the idea and reviewed the article.

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#### REFERENCES

- Aerts R, FS Chapin III. 1999. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *Advances in ecological research* 30: 1–68. DOI: <a href="https://doi.org/10.1016/S0065-2504(08)60016-1">https://doi.org/10.1016/S0065-2504(08)60016-1</a>
- Bahamonde HA, PL Peri, R Alvarez, A Barneix, A Moretto, G Martínez Pastur. 2013. Silvopastoral use of *Nothofagus* antarctica in Southern Patagonian forests, influence over net nitrogen soil mineralization. Agroforestry Systems 87: 259–271. DOI: https://doi.org/10.1007/s10457-012-9541-5
- Bahamonde HA, V Fernández, F Mattenet, PL Peri. 2016. Mineral elements in grasses growing in contrasting environmental conditions in southern Patagonia. *New Zealand journal of agricultural research* 59(3): 235-249. DOI: <a href="https://doi.org/10.1080/00288233.2016.1188132">https://doi.org/10.1080/00288233.2016.1188132</a>
- Bahamonde HA, G Martínez Pastur, MV Lencinas, R Soler, YM Rosas, B Ladd, S Duarte Guardia, PL Peri. 2018. The relative importance of soil properties and regional climate as drivers of productivity in southern Patagonia's *Nothofagus antarctica* forests. *Annals of forest science* 75(2): 1-8. DOI: https://doi.org/10.1007/s13595-018-0725-7.
- Bahamonde HA, D Sánchez-Gómez, J Gyenge, PL Peri, JM Cellini, I Aranda. 2019a. Thinking in the sustainability of *Nothofagus antarctica* silvopastoral systems, how differ the responses of seedlings from different provenances to water shortage? *Agroforestry Systems* 93: 689-701. DOI: <a href="https://doi.org/10.1007/s10457-017-0167-5">https://doi.org/10.1007/s10457-017-0167-5</a>
- Bahamonde HA, V Fernández, J Gyenge, F Mattenet, PL Peri. 2019b. Essential nutrient and trace element foliar resorption of two co-existing *Nothofagus* species grown under different environmental conditions in southern Patagonia. *Frontiers in Plant Science* 10: 1542. DOI: <a href="https://doi.org/10.3389/fpls.2019.01542">https://doi.org/10.3389/fpls.2019.01542</a>
- Diehl P, MJ Mazzarino, F Funes, S Fontenla, M Gobbi, J Ferrari. 2003. Nutrient conservation strategies in native Andean-Patagonian forests. *Journal of vegetation science* 14: 63–70. DOI: <a href="https://doi.org/10.1111/j.1654-1103.2003.tb02128.x">https://doi.org/10.1111/j.1654-1103.2003.tb02128.x</a>
- Diehl P, MJ Mazzarino, S Fontenla. 2008. Plant limiting nutrients in Andean-Patagonian woody species: effects of interannual rainfall variation, soil fertility and mycorrhizal infection. Forest Ecology and Management 255 (7): 2973-2980. DOI: https://doi.org/10.1016/j.foreco.2008.02.003
- Donoso C, L Steinke, A Premoli. 2006. *Nothofagus antarctica* (G. Forster) Oerst. *In* Donoso Zegers C Ed, Las especies arbóreas de los bosques templados de Chile y Argentina. Autoecología, Valdivia, Chile, p. 401–410.
- Farr TG, PA Rosen, E Caro, R Crippen, R Duren, S Hensley, M Kobrick, M Paller, E Rodriguez, L Roth, D Seal, S Shaffer, J Shimada, J Umland, M Werner, M Oskin, D Burbank, D Alsdorf. 2007. The shuttle radar topography mission. *Reviews of geophysics* 45(2): 1-33. DOI: <a href="https://doi.org/10.1029/2005RG000183">https://doi.org/10.1029/2005RG000183</a>
- Frangi JL, MD Barrera, LL Richter, AE Lugo. 2005. Nutrient cycling in *Nothofagus pumilio* forests along an altitudinal gradient in Tierra del Fuego, Argentina. *Forest Ecology and Management* 217(1): 80-94. DOI: <a href="https://doi.org/10.1016/j.foreco.2005.05.051">https://doi.org/10.1016/j.foreco.2005.05.051</a>

- Frangi JL, C Pérez, R Martiarena, M Pinazo, G Martínez Pastur, A Brown, P Peri, DS Ceballos. 2015. Aspectos ecológicos y ambientales de los bosques nativos y plantaciones forestales en la Argentina: Una visión panorámica y conceptual. *In* Casas RR, GF Albarracín Eds. El deterioro del suelo y el ambiente en Argentina. p. 365-432.
- Gallardo MB, C Pérez, M Núñez-Ávila, JJ Armesto. 2012. Desacoplamiento del desarrollo del suelo y la sucesión vegetal a lo largo de una cronosecuencia de 60 mil años en el volcán Llaima, Chile. Revista chilena de historia natural 85(3): 291-306. DOI: <a href="http://dx.doi.org/10.4067/S0716-078X2012000300004">http://dx.doi.org/10.4067/S0716-078X2012000300004</a>
- Gargaglione V, HA Bahamonde, PL Peri. 2018. Decomposition and nutrient release of grass and tree fine roots along an environmental gradient in southern Patagonia. *Austral Ecology* 44(2): 276-289. DOI: <a href="https://doi.org/10.1111/aec.12672">https://doi.org/10.1111/aec.12672</a>
- Hevia F, KL Decker, RE Boerner. 1999. Foliar nitrogen and phosphorus dynamics of three Chilean *Nothofagus* (Fagaceae) species in relation to leaf lifespan. *American Journal of Botany 86*(3): 447-455. DOI: <a href="https://doi.org/10.2307/2656765">https://doi.org/10.2307/2656765</a>
- Hijmans RJ, SE Cameron, JL Parra, PG Jones, A Jarvis. 2005. Very high-resolution interpolated climate surfaces for global land areas. *International Journal of Climatology* 25: 1965–1978. DOI: <a href="https://doi.org/10.1002/joc.1276">https://doi.org/10.1002/joc.1276</a>
- Ivancich H, G Martínez Pastur, PL Peri. 2011. Modelos forzados y no forzados para el cálculo de índice de sitio en bosques de *Nothofagus antarctica* en Patagonia Sur. *Bosque* 32: 135–145. DOI: <a href="http://dx.doi.org/10.4067/S0717-92002011000200004">http://dx.doi.org/10.4067/S0717-92002011000200004</a>
- Kabata-Pendias A. 2011. Trace elements in soils and plants. 4th ed. Boca Raton, FL: CRC Press.
- Kirkby E. 2012. Introduction, definition and classification of nutrients. *In Marschner's P Ed. Mineral Nutrition of Higher Plants*, 3rd Edition, p. 3-5. DOI: <a href="https://doi.org/10.1016/B978-0-12-384905-2.00001-7">https://doi.org/10.1016/B978-0-12-384905-2.00001-7</a>
- Lambers H, JG Bishop, SD Hopper, E Laliberté, A Zúniga-Feest. (2012). Phosphorus-mobilization ecosystem engineering: the roles of cluster roots and carboxylate exudation in young P-limited ecosystems. *Annals of botany* 110(2): 329-348. DOI: <a href="https://doi.org/10.1093/aob/mcs130">https://doi.org/10.1093/aob/mcs130</a>
- Marschner H, V Römheld. 1996. Root-induced changes in the availability of micronutrients in the rhizosphere. *In* Waisel Y, A Eshel, U Kafkaki Eds., Plant roots: the hidden half, 3rd edition, Marcel Decker, Inc., New York, p. 557-580.
- Peri PL, V Gargaglione, G Martínez Pastur. 2006. Dynamics of above- and below-ground biomass and nutrient accumulation in an age sequence of *Nothofagus antarctica* forest of

- Southern Patagonia. Forest ecology and management 233: 85–99. DOI: https://doi.org/10.1016/j.foreco.2006.06.009
- Peri PL, V Gargaglione, G Martínez Pastur. 2008. Above- and belowground nutrients storage and biomass accumulation in marginal *Nothofagus antarctica* forests in Southern Patagonia. *Forest ecology and management* 255(7): 2502–2511. DOI: https://doi.org/10.1016/j.foreco.2008.01.014
- Peri PL, V Gargaglione, G Martínez Pastur, MV Lencinas. 2010. Carbon accumulation along a stand development sequence of *Nothofagus antarctica* forests across a gradient in site quality in Southern Patagonia. *Forest ecology and management* 260: 229–237. DOI: <a href="https://doi.org/10.1016/j.foreco.2010.04.027">https://doi.org/10.1016/j.foreco.2010.04.027</a>
- Peri PL, SG Ormaechea. 2013. Relevamiento de los bosques nativos de ñire (*Nothofagus antarctica*) en Santa Cruz: base para su conservación y manejo. Ediciones INTA web. Accessed 28 feb. 2023. Available in <a href="https://repositorio.inta.gob.ar/handle/20.500.12123/11180#">https://repositorio.inta.gob.ar/handle/20.500.12123/11180#</a>
- Peri PL, Bahamonde, R Christiansen. 2015. Soil respiration in Patagonian semiarid grasslands under contrasting environmental and use conditions. *Journal of arid environments* 119: 1–8. DOI: <a href="https://doi.org/10.1016/j.jarid-env.2015.03.008">https://doi.org/10.1016/j.jarid-env.2015.03.008</a>
- Peri PL, MV Lencinas, J Bousson, R Lasagno, R Soler, H Bahamonde, G Martínez Pastur. 2016. Biodiversity and ecological long-term plots in Southern Patagonia to support sustainable land management: the case of PEBANPA network. *Journal of nature conservation* 34: 51–64. DOI: <a href="https://doi.org/10.1016/j.jnc.2016.09.003">https://doi.org/10.1016/j.jnc.2016.09.003</a>
- Pilon-Smits EA, CF Quinn, W Tapken, M Malagoli, M Schiavon. 2009. Physiological functions of beneficial elements. *Current Opinion Plant Biology* 12 (3): 267–274. DOI: <a href="https://doi.org/10.1016/j.pbi.2009.04.009">https://doi.org/10.1016/j.pbi.2009.04.009</a>
- Tabatabai MA, JM Bremner. 1991. Automated instruments for determination of total carbon, nitrogen, and sulfur in soils bycombustion techniques. *In* Smith KA Ed, Soil analysis modern instrumental techniques, 2nd ed. Marcel Dekker, New York, p. 261–286.
- Truong C, LA Gabbarini, A Moretto, JM Escobar, ME Smith. 2024. Ectomycorrhizal fungi and the nitrogen economy of Nothofagus in southern Patagonia. *Ecology and Evolution* 14(10): e70299. DOI: https://doi.org/10.1002/ece3.70299
- WorldClim. 2005. Global climate and weather data. Accessed 31 mar. 2024. Available in <a href="https://www.worldclim.org">https://www.worldclim.org</a>
- Zhou J, A Zúñiga-Feest, H Lambers. 2020. In the beginning, there was only bare regolith—then some plants arrived and changed the regolith. *Journal of Plant Ecology* 13(5): 511-516. DOI: https://doi.org/10.1093/jpe/rtaa030

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