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Variations in the hierarchical architecture of *Nothofagus obliqua*: evidence from juvenile naturally-regenerated trees and from seedlings obtained by controlled crosses of selected individuals

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Abstract

Background: Many tree species with potential for wood production present high intra-specific variations in the extent of trunk differentiation (i.e. hierarchical architecture). The identification of multiple-scale traits related to hierarchical architecture could improve selection criteria for domestication.

Methods: We investigated the hierarchical architecture of *Nothofagus obliqua*, a valuable species for timber production, but with high structural variability. Young trees in even-aged natural regeneration gaps, and seedlings derived from controlled crosses and open pollination were studied. For the second approach, trees with contrasting degree of hierarchical architecture were manually crossed. In both, juvenile plants and seedlings, we analyzed trunk growth unit traits and hierarchical architecture indices based on the relative size and branching angle of main branches.

Results: In regeneration gaps, hierarchical architecture was positively related to height and diameter. Apex persistence, the number of sylleptic branches and mean internode length were indicative of larger and more hierarchical trees. Some support is provided to the idea that adult trees with a notably hierarchical architecture could produce young trees with early signs of a hierarchical architecture. In seedlings, hierarchical architecture was negatively related to basal diameter.

Conclusions: Some growth unit traits that differed among progenies and were related to hierarchical architecture could be considered for the development of selection criteria for young trees.

Keywords: architecture; controlled-crosses; hierarchy; domestication; annual shoot

Introduction

Unravelling the way in which tree architecture is affected by genetic and environmental factors (Barthélémy & Caraglio 2007) is a major goal of programs aimed at the selection and domestication of species for timber production. Such studies attempt to advance in the identification of factors involved in the expression of wanted and unwanted architectural traits (Puntieri et al. 2013b; Vargas-Reeve et al. 2013). A major step forward in this regard would be the detection of traits that, being expressed by individuals at early ontogenetic stages (seedlings and saplings), are correlated with architectural traits at adult stages. Within this context, it is necessary to consider the modular structure of plants, and to assess the relationship between morphological and growth patterns at different scales, from metamers (i.e. the smallest structural unit of a plant, made up by an internode and a node-with its associated leaf/leaves and axillary bud/s) to whole trees. This multi-scale approach has been applied in intra-specific comparisons between progenies and provenances (Puntieri et al. 2013b; Soliani & Aparicio 2020).

Trees cultivated for high-quality timber production are sought to develop a strongly hierarchical architecture, with a single, straight and dominant trunk, clearly differentiated from its lateral branches by both size and spatial orientation (Ford 1985; Cameron et al. 2012; Vargas-Reeve et al. 2013). The development of large and persistent branches with an almost vertical orientation, codominant with the trunk (like in trunk forks), is an undesirable feature for timber production (Drénou et al. 2020). The correlation between traits that characterize a hierarchical crown architecture, and fine-scale traits such as those of annual shoots - that may be observed in seedlings and saplings - could be useful for the early selection of trees. Correlations between the morphologies of shoots and trunk have been studied in tree species (Osada 2011) but, to our knowledge, not from the perspective of tree selection for domestications purposes. In this regard, among the potentially useful traits of annual shoots are the length, diameter and number of nodes of the stem, branching pattern, and apex persistence, all of which are indicative of axis vigour in several temperate tree species (Remphrey & Powell 1984; Puntieri et al. 1998), and could be related to hierarchical structure (Marron et al. 2006). In addition, the angles formed between main branches and the trunk are indicative of the likelihood of trunk forking. Part of the variation of this trait have been shown to be genetically determined in coniferous species (Cameron et al. 2012), making it a potentially selectable trait. The persistence over time of the trunk's apex, which also could exhibit genetic variations (Puntieri et al. 2007), may be relevant as an indicator of the extent of endurance of the trunk as the dominant vertical axis of a tree; nevertheless, the relation of this trait with the hierarchical structure of trees has not been analyzed so far. Improving the current knowledge about genetic variations of morpho-architectural attributes in tree species with productive potential and high architectural variability may contribute to shorten the selection cycles and increase the diversity of production systems.

Several tree species from temperate forests of Patagonia have suitable characteristic for wood production, particularly those included within the genus *Nothofagus,* whose natural populations have been severely exploited in Argentina and Chile during the 20th century (Marchelli et al. 2020). In the last decades, significant advances have been made towards the domestication of some *Nothofagus* species, but their cultivation is scarce and wood is still obtained from natural populations (Marchelli et al. 2020). Among the challenges facing the domestication of these and other native species from Patagonia, is the selection of individuals with a strongly hierarchical structure and the assessment of factors related to structural variations. One of the most promising *Nothofagus* species for domestication programs is *N. obliqua* (Mirb.) Oerst., because of its high potential for wood production (Donoso et al. 2006; Azpilicueta et al. 2020). Argentinean populations of this species conserve high genetic diversity (see Azpilicueta et al. 2020), which may be the source of morphological and architectural variability within and between populations upon which domestication programs may operate (Puntieri et al. 2013a; Corvalán Vera 2017). Previous studies have attempted to assess genetic and environmental factors affecting architectural traits in *Nothofagus* spp., by means of common-garden trials including open-pollinated progenies obtained from different provenances (e.g. Puntieri et al. 2006; Puntieri et al. 2013a; Aparicio et al. 2015). Such studies could be complemented by others analyzing progenies derived from the controlled crossing of individuals selected based on their architectural features. Nevertheless, to our knowledge, progenies of *Nothofagus* in which both parental origins were previously selected are not available so far. A major limitation for performing controlled crosses is the large inter-annual variation in flower production observed in individuals of several *Nothofagus* species (Torres et al. 2016); the synchronous occurrence of high flower production in selected trees could be very infrequent.

In this work we analyzed architectural traits related to hierarchical growth, considering different scales of analysis and assessing variations between progenies. In *N. obliqua,* a tree species with high potential for timber production from southern forests of Argentina and Chile, this approach attempts to identify useful traits for the development of early selection criteria of individuals with desirable architecture for cultivation*.* The plant material used consisted of juvenile individuals from natural regeneration in a managed forest, and seedlings obtained either by controlled crosses, or by open pollination of adult individuals previously selected according to their architecture. In the former case, the hierarchical development of juvenile trees was linked to trunk size and to the morphology of the most recent annual shoots that made up the trunk. The study of progenies from controlled crosses made it possible to evaluate to what extent the hierarchical structure of the parental individuals affects that of their offspring.

Methods

Hierarchical development in naturally-regenerated juvenile plants

Measurements were performed in plants belonging to a natural population located at Cerro Quilanlahue (Lanín National Park, 40° 8' S, 71° 28' E; 900 m a.s.l.). The study site covers an area of 3 ha within a mixed forest of *N. dombeyi*, *N. nervosa* and *N. obliqua* (Sola et al. 2016). At the beginning of the 1990s, forest management was performed in this area through the thinning of adult trees so as to reduce canopy cover to about 30-40%. Tree regeneration in this area takes place in patches (Sola et al. 2015). For the present study, nine naturally-regenerated patches of *N. obliqua*, each one between 50-100 m^2 in area, were selected in 2014. At the time of measurement, vegetation cover in each patch varied between 72 and 80% (estimated through four hemispheric photographs per patch, taken at 2 m from the ground). Based on the observation of morphological marks delimiting successive annual shoots of the trunk (following criteria previously applied in *Nothofagus* spp; e.g. Magnin et al. 2016), 10-15 year-old individuals (mean age=12 years) were selected. One hundred trees were finally obtained from the nine regeneration patches (one patch provided 20 individuals, and the others provided 10 individuals each). For each individual, the distances to the six closest neighbor trees were measured and then an average value was computed. Morphological measurements performed on each individual included two scales of analysis: whole tree, and the trunk portion extended in the last two growing seasons, that is, its two most distal growth units (GU: axis segment extended during a single flush). The main branch with the largest basal diameter arising directly from the trunk (hereafter MB) was identified in each tree.

Traits considered at the whole-tree scale were: tree age, height, trunk diameters at ground level and at the site of insertion of the MB ($Trunk_d$), basal diameter of the MB (MB_d), and the divergence angle formed between the MB and the trunk (MBλ; Fig. 1). Tree heights were measured with a measuring tape to the nearest 1 cm, diameters with digital calipers (0.01mm precision) and angles with a protractor. For the second scale of analysis, the two most recent GU of the trunk (current trunk-GU and 2-yr trunk-GU), and the thickest lateral GU arising from the 2-yr trunk-GU (current lateral GU; Fig. 1) were identified and their respective basal diameters measured. For the current trunk-GU, apex condition (persistent or dead), stem length, number of nodes and number of sylleptic branches (i.e. branches developed

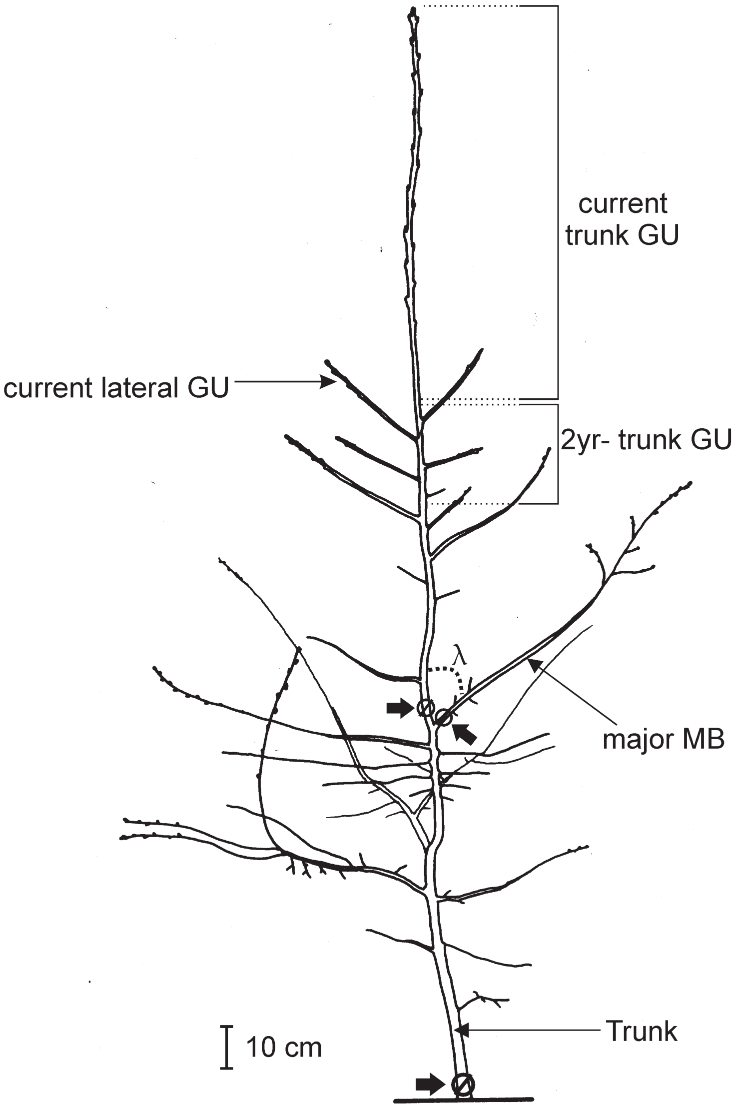

FIGURE 1: Semi-schematic representation of a juvenile *N. obliqua* tree. The variables measured at the scale of the whole plant and at the scale of the last two years of trunk growth are indicated. Ø= sites of stem diameter measurement; λ= angle. GU= growth unit; MB= major main branch.

simultaneously with its bearing GU) were registered. Stem slenderness was computed as stem length/stem diameter, and mean internode length as stem length/ number of nodes, for each tree.

Variables adopted as indicators of a hierarchical tree structure were:

(1) $Trunk_d$/ MB_d ratio and

(2) the MBλ (Table 1).

We considered that a more hierarchical structure would be related to a high value of $Trunk_d/MB_d$ ratio and a broad MBλ. For the distal portion of each tree, the degree of hierarchical development was assessed by means of the ratio between the diameter of the current trunk GU and the diameter of the current lateral GU; this ratio would be positively related to hierarchical development (Table 1).

Hierarchical structure of plants derived from controlled crosses

In winter 2015, four adult trees of *N. obliqua* were selected from a natural population located at Lanín National Park, Argentina (40°08’35” S, 71°37’42” W; 650 m a.s.l.). Two of the selected individuals (hereinafter *H1* and *H2*) presented what we evaluated as a highly hierarchical structure: a straight trunk well differentiated from its lateral branches and without forks. The other two individuals (hereinafter *NH1* and *NH2*) presented a less hierarchical architecture; they had at least three clearly identifiable trunk forks and several large branches. By means of manual pollination (following Torres & Puntieri 2013), the following crosses were performed: *H1xH2*, *H1xNH2*, *NH1xH2* and *NH1xNH2* (*ovule donor x pollen donor*). At the end of the growing season, the seeds derived from the pollinated flowers as well as seeds derived from open pollination were collected from each ovule donor. Seed germination and seedling treatments were performed according to the protocol proposed by Schinelli Casares (2012). The obtained seedlings (Table 2) were kept in a glasshouse at *Instituto Nacional de Tecnología Agropecuaria*, Trevelin, Argentina (43°07’17” S, 71°33’42” W; 369 m a.s.l.) for one growing season. Afterwards, the plants were moved outdoors in the same nursery and distributed along six rows, located 30 cm apart from each other. The future transport of these seedlings to a field trial was projected, so that plants were grouped by family, as it was considered appropriate from a logistical viewpoint. Water was provided regularly and light availability could be assumed to be similar for all progenies, since no large trees or buildings that could cause shading were present surrounding the essay; therefore, we assumed that environmental variations related to plant distribution were negligible.

Morphological measurements of seedlings were performed at the end of each of two successive growing seasons (2017-2018 and 2018-2019) corresponding to the second and third growing seasons of these seedlings. Henceforth, GU1, GU2 and GU3 will be used to refer to the trunk GU developed during the first (2016-2017), second (2017-2018) and third (2018-2019) growing seasons, respectively. Stem length, basal diameter and number of nodes (= number of leaves) were measured for GU1, GU2 and GU3. The basal diameter of each plant was registered at the end of the third growing season. The following variables describing the hierarchical structure of the main axis were registered per plant:

(1) hierarchy indexes (H_{index}), computed as: $H_{indexGU2}$= diameter GU2/diameter of the major branch arising from GU1; and $H_{indexGU3}$= diameter GU3/ diameter of the major branch arising from GU2;

(2) insertion angle of the major branch arising from GU1 and GU2 (MBλ), considering a qualitative scale from narrow to wide angles: MBλ < 22.5°, 22.5° ≤ MBλ < 45°, 45° ≤ MBλ < 67.5°, 67.5° ≤ MBλ;

TABLE 1: Variables describing growth and hierarchical development at different scales, measured in juvenile plants of *N. obliqua*. Abbreviations (code) used in the text are also shown.

Trait	Variable	Code
Tree size and morphology		
	Tree height	-
	Basal trunk diameter	-
	Stem slenderness (height/diameter ratio)	-
Tree hierarchy		
	Trunk diameter at MB insertion/MB basal diameter	Trunkd/MBd ratio
	MB insertion angle relative to trunk	MBλ
Current trunk-GU morphology and hierarchy	Current trunk-GU diameter/current lateral-GU diameter	TrunkGUd/branchGUd ratio
	Apex condition of current trunk GU	-
	number of sylleptic branches on current trunk GU	-
	Length/diameter	Stem slenderness
	Length/Number of nodes	Mean internode length

MB= major main branch; GU= growth unit

TABLE 2: Number of seedlings obtained from each progeny, after three growing seasons.

Ovule donor	Pollen donor	Crossing code	N° seedlings
H1	H2	H1 x H2	32
	NH2	H1 x NH2	56
	Open pollination	H1 open pollination	55
NH1	H2	NH1 x H2	75
	NH2	NH1 x NH2	107
	Open pollination	NH1 open pollination	50

(3) number of sylleptic branches produced by GU2 and GU3;

(4) apex condition of GU2 and GU3.

$H_{indexGU2}$ and $H_{indexGU3}$ were both measured at the end of the third growing season, so that one year of secondary growth was included in the measure of $H_{indexGU2}$, but not in the measure of $H_{indexGU3}$

Statistical analyses

For the juvenile plants of the natural population and at the whole tree scale, we studied the relationship between plant vigour and hierarchical development. Linear mixed models were applied to test variation in (1) height and (2) basal diameter as functions of $Trunk_d/MB_d$ ratio and MBλ. As both variables are correlated (r=0.33, p<0.001), we tested their effects in separate models. All tested models also considered age and mean distance to neighbor trees as predictors (both fixed), whereas the patch was included as a random factor. Therefore, the following mixed-effects models were tested:

$$Y_{ij} = (\beta_0 + \mu_{0j}) + \beta_1 * age_{ij} + \beta_2 * mn_{ij} + \beta_3 * (Trunk_d/MB_d)_{ij} + e_{ij} \quad (1)$$

$$Y_{ij} = (\beta_0 + \mu_{0j}) + \beta_1 * age_{ij} + \beta_2 * mn_{ij} + \beta_3 * MB\lambda_{ij} + e_{ij} \quad (2)$$

In both cases, $e_{ij} \sim N(0, \sigma^2)$ being mutually independent

In both models, Y_{ij} represents the value of the response variable (height or diameter) of the *i* individual (1,..., 100) nested in the *j* regeneration patch (1,..., 9; random); β_0 represents the global intercept, μ_{0j} represents the random intercept (i.e. the mean deviation from the overall mean for the *j* regeneration patch), and e_{ij} represents the residual error; where: *mn* represents mean distance to neighbors; *MBλ*, insertion angle of the major main branch; $Trunk_d/MB_d$, ratio between trunk diameter and *MB* diameter. Variables were log-transformed whenever necessary to conform with homoscedasticity and/or normality assumptions. In all selected models, fixed effects were tested by means of analysis of deviance type III Wald F-tests with Kenward-Roger degrees of freedom (Kenward & Roger 1997).

In order to assess the degree to which the hierarchical structure and morphology at the scale of current trunk-GU were related to trunk vigour, morphology and hierarchy, mixed-effects models with the following structure were applied:

$$Y_{ij} = (\beta_0 + \mu_{0j}) + \beta_1 * age_{ij} + \beta_2 * mn_{ij} + \beta_3 * height_{ij} + \beta_4 * trunk\ basal\ diam_{ij} + \beta_5 * trunk\ slenderness_{ij} + \beta_6 * (Trunk_d/MB_d)_{ij} + \beta_7 * MB\lambda_{ij} + e_{ij} \quad (3)$$

$e_{ij} \sim N(0, \sigma^2)$ being mutually independent

For the *i* individual (1,..., 100) nested in the *j* regeneration patch (1,..., 9; random), Y_{ij} represents one of the following traits related to hierarchy at the scale of the current trunk annual shoot: (1) $TrunkGU_d/branchGU_d$ ratio, (2) GU stem slenderness, (3) number of sylleptic branches, (4) apex condition and (5) mean internode length. β_0 represents the global intercept, μ_{0j} represents the random intercept. In the case of apex persistence, a binary trait that can take only two values (live vs death apex), models used considered a binomial distribution and a logistic link function. For the count trait number of sylleptic branches, the applied model assumed a negative binomial distribution, with a logarithmic link function. For each model fitted to continuous traits, the significance of the effect of each fixed variable (variables 1, 2 and 5) was tested by means of ANOVA type III Kenward-Roger (Kenward & Roger 1997). Chi-square tests of sequential deviance were applied to evaluate models corresponding to non-normally distributed variables (3 and 4).

For all of the above tests, model selection was performed by starting with the full model and then discarding (1) variables unrelated to the response variable and (2) redundant variables; a reduced model was then selected with a minimum set of predictors and maximum explanation of the response variable. All mixed models were fitted with the 'lmer' function from the 'lme4' package (Bates et al. 2015) in R 3.4.2 (R Development Core Team 2024).

Comparisons between progenies obtained by controlled crosses and open pollination were performed by means of factorial models (2 ovule donors x 3 pollen origins). The compared variables were: plant basal diameter after three growing seasons, stem slenderness and mean internode length for GU2 and GU3. Only GU2 and GU3 were included in this analysis since GU1 were developed under greenhouse conditions. Variations concerning $H_{indexGU2}$ and $H_{indexGU3}$ were analyzed by means of a model considering plant basal diameter, ovule origin and pollen origin; the interaction between ovule and pollen origin was also tested. The other variables related to hierarchical structure were analyzed by means of Generalized Linear Models; the considered distribution of each model depended on each variable: a multinomial model was tested for $MB\lambda$, which can take four discrete values; negative binomial distribution (logarithmic link function) for the number of sylleptic branches per GU (count variable; Poisson model was discarded because of over-dispersion), and binomial distribution (logistic link function) for the probability of apex persistence (binary trait). For those models concerning non-normal data distributions, Chi-square tests of sequential deviance were applied, including the covariable corresponding to each model: branch diameter in the case MBλ, and plant basal diameter for all other variables.

All statistical analyses were performed with the R software (R Development Core Team 2024); a 5% significance level was adopted.

Results

Variations in the hierarchical structure of juvenile plants in natural conditions

$Trunk_d/MB_d$ and $MB\lambda$ were positively related to tree height (Fig. 2a, b) and to trunk basal diameter (Fig. 2c, d). The $Trunk_d/MB_d$ ratio presented stronger relationships with tree height (F= 13.2, $p<0.001$) and basal diameter (F=15.6, $p<0.001$) than $MB\lambda$, (F=7.1, p=0.01; F=6.1, p=0.01, respectively) after accounting for plant age and mean neighbor distance.

When considering the distal end of the trunk, the $TrunkGU_d/branchGU_d$ ratio presented a positive relationship only with plant basal diameter (F=5.6, p= 0.02), and a slightly negative relationship with plant age (F=4.0. p=0.05; Table 3). The stem slenderness of the current trunk-GU was not significantly related to any of the variables describing vigour and hierarchical structure at the whole plant scale, whereas the persistence of its apex was more frequent in plants with a thicker trunk (χ^2=18.2, $p<0.001$) and with higher values for the $Trunk_d/MB_d$ ratio (χ^2=7.8, p=0.005; Table 3). Plants with higher $MB\lambda$ tended to present higher number of sylleptic branches on the current trunk-GU (χ^2=4.5, p=0.03; Table 3). Mean internode length of the current trunk-GU was positively related to plant height and diameter, which were correlated to one another; the best-fit model was the one that included plant height as predictor variable of mean internode length (F=17.8, $p<0.001$; Table 3).

Architectural development of plants obtained by controlled crosses

After three growing seasons, plants of all progenies presented similar mean basal diameters (F_{ovule}=2.7, F_{pollen}=1.3, $F_{ovule*pollen}$=2.9; in all cases $p>0.05$). Variation in the slenderness of GU2 depended on the interaction between ovule origin and pollen origin ($F_{ovule*pollen}$=6.6, $p<0.01$). This trunk portion was, on average, thicker for plants derived from the *NH1 x NH2* crossing than for plants of all other crosses except *NH1 x H2* (Fig. 3a). The stem slenderness of GU3 varied with pollen origin (F_{pollen}=4.5, p=0.01), while the effect of ovule origin was marginal (F_{ovule}=3.4, p=0.06); the interaction between pollen origin and ovule had no effect on this variable ($F_{ovule*pollen}$=1.7, p=0.20). This trunk portion tended to have a slenderer stem in plants derived from the pollination with *H2*, than in plants from other pollinations, but such a trend was statistically confirmed only for the *H1* progeny (Fig. 3a). Mean internode length was affected by the interaction between both parental origins for GU2 ($F_{ovule*pollen}$=10.8, $p<0.001$) and GU3 ($F_{ovule*pollen}$=3.6, p=0.03). Among the progeny of ovule donor *H1,* variations related to pollen origin were not consistent between GU2 and GU3 (Fig. 3b). For *NH1* progeny, variations due to pollen origin

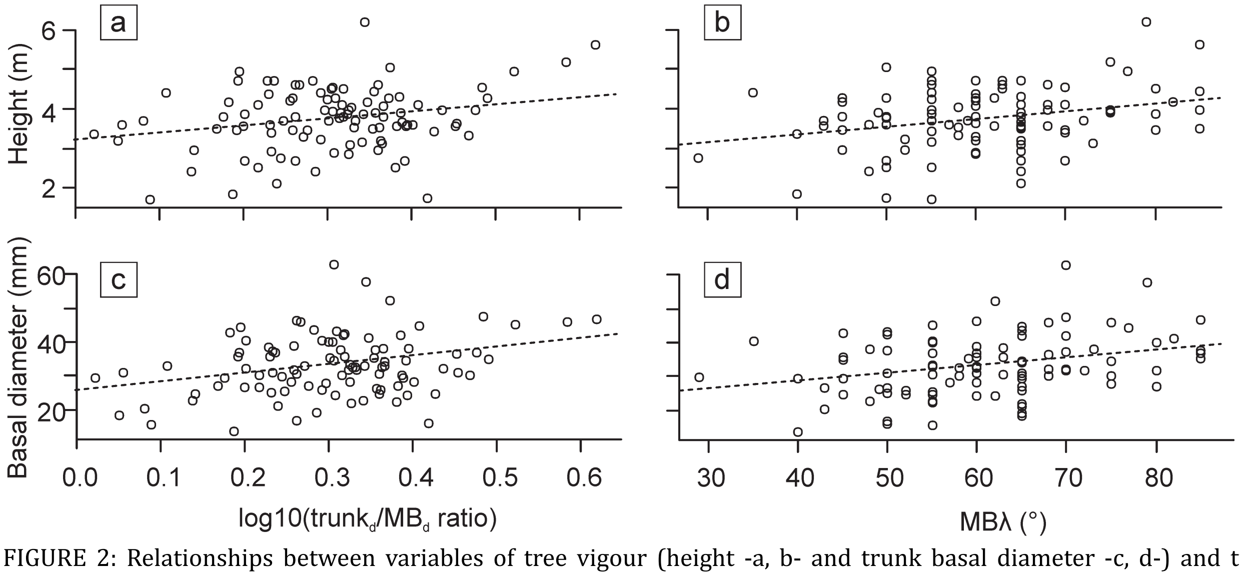

FIGURE 2: Relationships between variables of tree vigour (height -a, b- and trunk basal diameter -c, d-) and traits associated with hierarchical architecture ($Trunk_d/MB_d$ ratio and $MB\lambda$) in juvenile plants of *N. obliqua*. Linear-regression lines are shown. $Trunk_d/MB_d$ ratio= trunk diameter at the site of insertion of the major main branch (*MB*) divided by the basal diameter of the *MB*; $MB\lambda$= angle formed between the *MB* and the trunk.

TABLE 3: Models relating current trunk-GU traits with mean neighbor distance, plant age, plant size (height and basal diameter), stem slenderness (trunk height/diameter ratio) and hierarchical structure (variables in bold type).

Tree attribute *(x)*	**Current trunk GU *(y)***				
	TrunkGU$_d$/ branchGU$_d$^{1*}	**Trunk-GU stem slenderness^{1*}**	**Trunk-GU apex persistence^{2*}**	**Trunk-GU N° sylleptic branches^{3*}**	**Trunk-GU internode length^{1*}**
Intercept	1.44(0.22)	11.46(1.82)	-8.59(2.08)	1.21(0.85)	1.40(0.25)
Mean neighbour distance	ns	ns	0.005(0.003)	ns	ns
Plant age	-0.04(0.02)	ns	ns	ns	ns
Height	ns	ns	ns	ns	0.23(0.05)
Basal diameter	0.01(0.00)	ns	0.32(0.11)	ns	ns
Stem slenderness	ns	ns	ns	ns	ns
***Trunk$_d$/MB$_d$* ratio**	ns	ns	1.74(0.65)	ns	ns
MBλ	ns	ns	ns	0.02(0.008)	ns

The estimators of the parameters (standard error) corresponding to each variable with a significant effect ($p<0.05$) on the response variable are shown (see text for statistical and significance values). ns= parameter not significantly different from 0. *TrunkGU$_d$/branchGU$_d$* =current trunk-GU diameter/current lateral-GU diameter; Trunk-GU stem slenderness=length/diameter of the stem of the trunk GU; *Trunk$_d$/MB$_d$* ratio=trunk diameter at main branch insertion/main branch basal diameter; *MBλ*= main branch insertion angle relative to trunk.

^{1*}model with Normal distribution; ^{2*} Binomial model, parameters in logit-scale; ^{3*} Negative Binomial model, parameters in logarithmic scale.

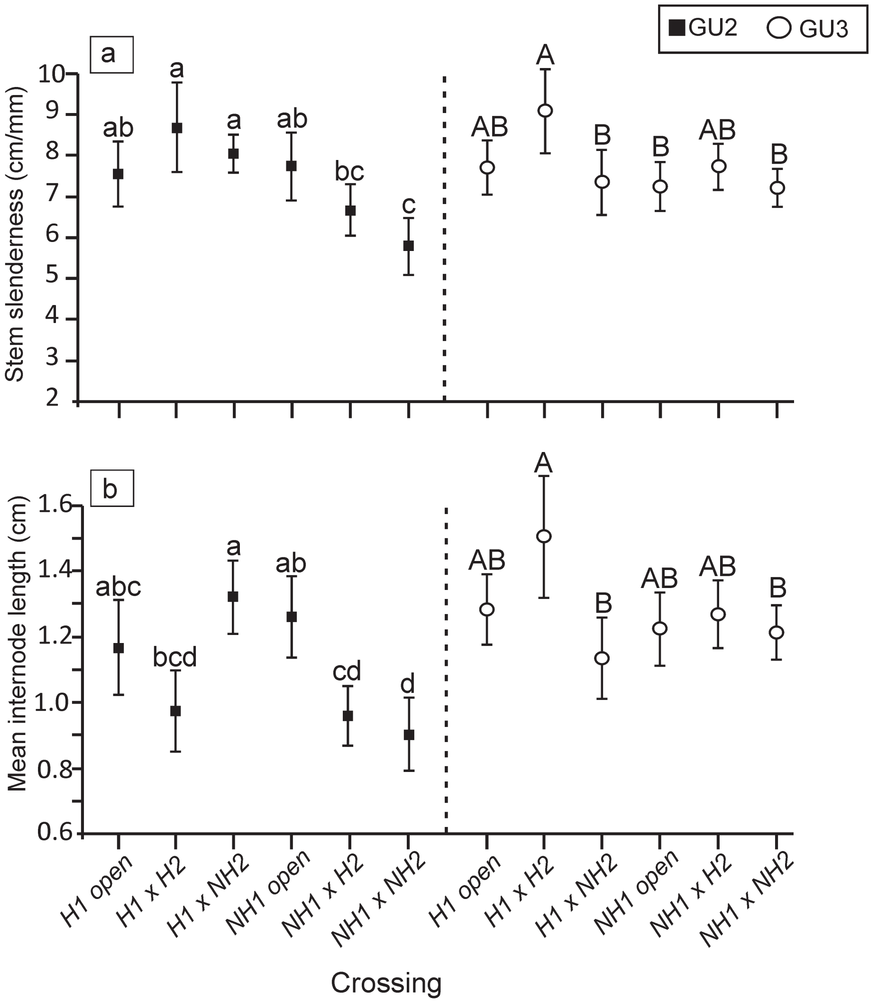

FIGURE 3: Mean (±95% confidence interval) values of stem slenderness (length/diameter; a) and mean internode length (b) for the growth units (GU) developed in the second and third growing seasons (GU2 and GU3 respectively) of *N. obliqua* plants derived from open pollination and from controlled crossings. *H1* and *NH1*: ovule-donor trees; *H2* and *NH2*: pollen-donor trees. *H* and *NH*: trees with hierarchical and non-hierarchical architecture, respectively. Statistical differences between progenies concerning GU2 are indicated with lowercase letters, whereas differences concerning GU3 are indicated with capital letters.

were evident only for GU2; where seedlings derived from open pollination had longer internodes than those derived from controlled cross-pollinations. In the case of GU3, plants from both ovule origins derived from the pollination with *H2* tended to present longer internodes than those derived from the pollination with *NH2*; this trend was statistically supported only for the *H1* progeny (Fig. 3b).

The hierarchy index corresponding to GU3 presented significant variations only between plants derived from different ovule donor (F=4.4, p=0.04), being higher in plants of the *H1* progeny (Fig 4a). In the case of GU2, the hierarchy index measured at the end of the third growing season did not vary among progenies, and was negatively related to the basal diameter of plants (F=67.4, p<0.001; Fig. 4c).

Thicker main branches tended to form narrower angles with GU1 and GU2 of the trunk than thinner ones (Table 4). Variations in MBλ between plants were related to ovule origin but not to pollen origin (Table 4). Plants of the *H1* progeny presented a higher frequency of narrow MBλ (lower than 22.5°) and a lower frequency of intermediate MBλ (between 22.5° and 67.5°) on GU1 than plants of the *NH1* progeny; the frequency of MBλ higher than 67.5° was low for both ovule origins. Low frequencies of narrow MBλ were observed for GU2. MBλ between 22.5° and 45° were more frequent for plants of the *H1* progeny, whereas those of the *NH1* progeny presented higher frequencies of MBλ higher than 45° (Fig. 5).

The number of sylleptic branches on the trunk was positively related to GU diameter and was significantly affected by the interaction between ovule origin and pollen origin (Table 4; Fig. 6). Differences between plants derived from different pollen origin were more notable for the *H1* progeny, although such variations were not consistent for GU2 and GU3. For this progeny, plants derived from open pollination developed less sylleptic branches in GU2 than plants derived from *H2* and *NH2;* for GU3, more sylleptic branches were produced by plants derived from the pollination with *H2* as compared to the progenies from other pollen origins. The number of sylleptic branches per GU was not related to pollen origin in the case of the *NH1* progeny.

For the GU developed in the second growing season (GU2), apex persistence was significantly more frequent for the *H1* progeny (in 65% of plants) than for the *NH1* progeny (43%; Table 4); no variations among pollen origins were confirmed (Table 4). At the end of the third growing season, apex persistence was positively related to the basal diameter of GU3, and varied significantly among progenies of different pollen origin (Table 4). In the case of the *H1* progeny, higher proportions of apex persistence were observed for plants derived from *H2*

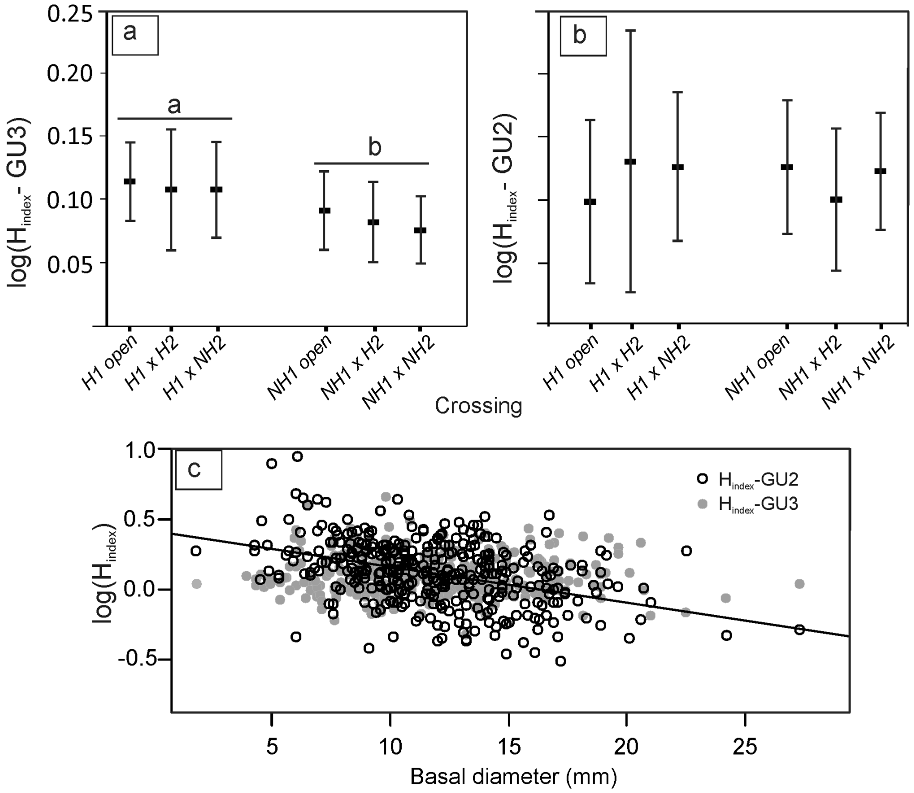

FIGURE 4: (a and b) mean (±95% c.i.) values of the hierarchy index computed for each progeny for two successive growing seasons. H_{index} -GU3= diameter GU3 / diameter of major branch arising from GU2; H_{index} -GU2= diameter GU2 / diameter of major branch arising from GU1. Different letters on top of the error bars indicate significant differences. (c) relationship between basal diameter of the trunk and H_{index} computed for GU2 and GU3; fit line shown for H_{index}-GU3, for which a significant relationship was found.

TABLE 4: Chi-square analyses of sequential deviance comparing models assessing the variation of the growth unit traits among 3-year plants of different genetic origin regarding ovule and pollen donor. Starting from the null model, AIC, residual deviance, model degrees of freedom and significance value for the sequential inclusion of each variable are shown.

		Trunk GU1				**Trunk GU2**			
Y	**Model**	**AIC**	**Res. Dev.**	**DF**	**p**	**AIC**	**Res. Dev.**	**DF**	**p**
MBλ	Null	767.8	761.8	3	-	892.2	886.2	3	-
	MB basal diameter	756.2	744.5	6	***	847.2	835.2	6	***
	ovule donor	749.1	731.1	9	***	841.9	823.9	9	0.010
	pollen donor	749.0	719.5	15	ns	849.9	819.9	15	ns
	ovule*pollen	755.7	713.7	21	ns	858.7	816.7	21	ns
		Trunk GU2				**Trunk GU3**			
Sylleptic branches	Null	973.7	499.5	2	-	1225.0	759.2	1	-
	GU diameter	863.7	307.4	3	***	999.8	327.1	2	***
	ovule donor	865.1	306.8	4	ns	1001.0	326.3	3	ns
	pollen donor	861.3	298.7	6	0.018	1004.6	325.9	5	ns
	ovule*pollen	856.2	289.4	8	0.009	1002.3	319.1	7	0.04
Shoot apex persistence	Null	455.9	453.9	1	-	519.1	517.1	1	-
	GU diameter	454.7	450.7	2	ns	366.9	362.9	2	***
	ovule donor	441.1	435.1	3	***	366.7	360.7	3	ns
	pollen donor	441.2	431.2	5	ns	361.8	351.8	5	0.011
	ovule*pollen	444.2	430.2	7	ns	362.0	349.0	7	ns

MBλ=insertion angle of the major main branch- Multinomial model; number of sylleptic branches- Negative Binomial model; Shoot apex persistence- Binomial model. *$p<0.05$, **$p>0.01$, ***$p<0.001$, ns, not significant.

and *NH2* (53 and 55% respectively) than for plants derived from open pollination (47%). For the *NH1* progeny, the proportion of GU3 with persistent apex was highest in plants derived from pollination with *H2* (60%), intermediate in plants derived from pollination with *NH2* (46%) and lowest in those derived from open pollination (36%).

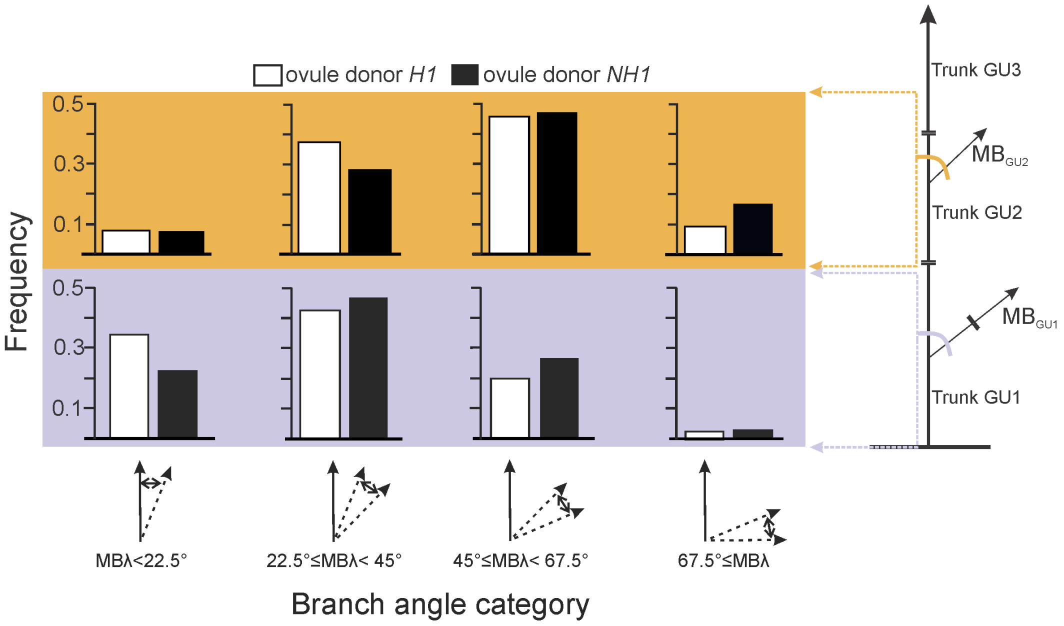

FIGURE 5: Frequency of each angle of insertion of the major main branch (MB) on the trunk for trunk growth units developed in the first (GU1) and second (GU2) growing seasons. Data corresponding to two ovule donors are indicated in white bars (H1, ovule donor with hierarchical structure; N=143) and black bars (NH1, ovule donor with non-hierarchical structure; N=232).

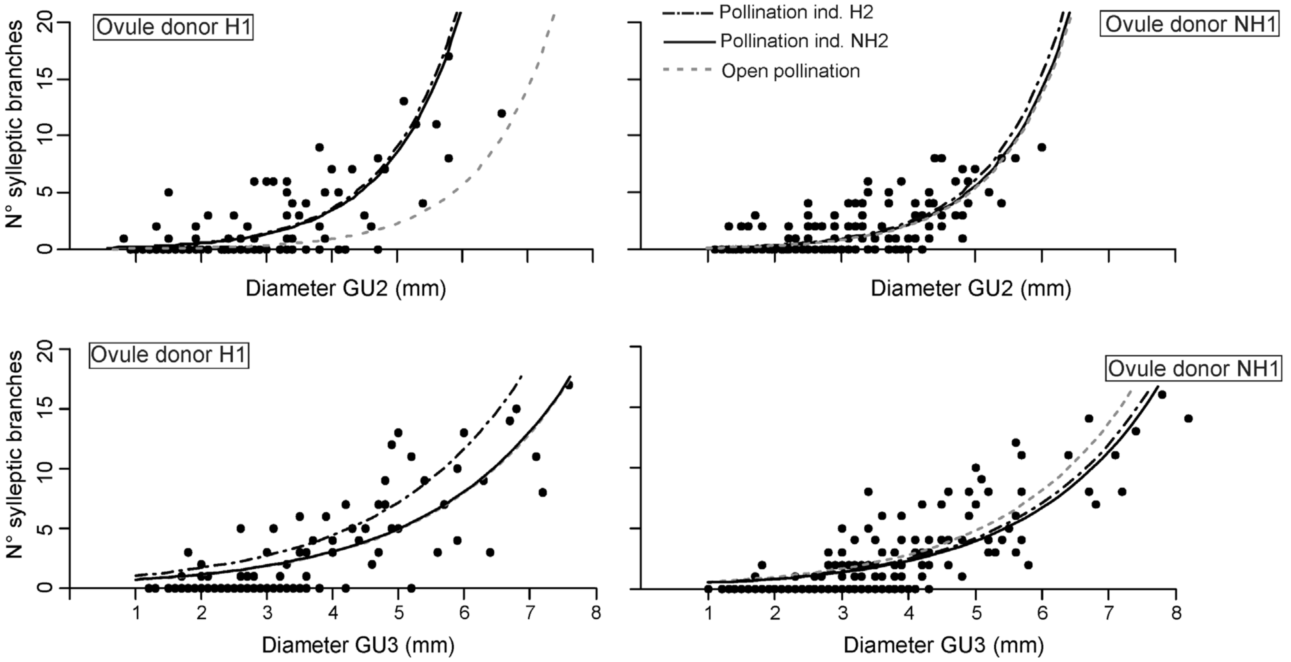

FIGURE 6: For plants of each progeny, models relating the number of sylleptic branches and stem diameter, for trunk growth units of two successive growing seasons (GU2 and GU3).

Discussion

Growth and hierarchical structure in natural regeneration patches

In even-aged regeneration patches of *N. obliqua*, more vigourous trees tended to have a more hierarchical architecture than less vigourous trees. This conclusion was supported by the fact that higher trunk diameter and height were associated with (1) lower diameter of the major branch relative to that of the trunk, and (2) a wider angle between the major branch and the trunk. These results highlight the role of the hierarchical growth in inter-individual competition to reach the canopy, as found for other deciduous species with moderate to low shade-tolerance (Millet et al. 1999). Contrary to this pattern, negative correlations between tree vigour and architectural hierarchy have been observed in some conifer species, in which trees with high growth rates in height and volume are more likely to develop trunk forks (Cumbie et al. 2012; Xiong et al. 2014). These contrasting results suggest that the correlation between vigour and hierarchical architecture could present inter-specific variations, maybe due to autoecological and/or morphological traits.

Our results showed that some of the traits of trunk GU could be related to hierarchical architecture. In this regard, apex persistence after GU extension was found to be more likely in trees with a thicker trunk and a high Trunk$_d$/MB$_d$ ratio than in those with opposite characteristics. The spontaneous death of the shoot apex during extension is often observed in *Nothofagus* spp. (Barthélémy et al. 1999; Puntieri et al. 2007) and many other tree species (Drénou 2000). In the case of *Nothofagus*, further extension of the concerned axis relies on the development of a shoot from a sub-apical bud so that the death of the apical meristem of a trunk shoot may not affect its long-term straightness (Puntieri et al. 2007). Nevertheless, our results indicate that the structural hierarchy of the trunk is related to shoot-apex persistence. A higher susceptibility to shoot-apex death could mean a weaker apical control (Drénou 2000), which could allow hormonal and nutritional inputs for the development of a vigourous main branch, codominant to the trunk (Cline & Harrington 2007). The endogenous variations in apex persistence shown for *N. obliqua* (Puntieri et al. 2007; present study), support the idea that this trait could be a useful criterion for the selection of hierarchical trees at early ontogenetic stages.

Plants with wider angles between the major main branch and the trunk presented a higher number of sylleptic branches than those with a narrow-angled major branch. Sylleptic branches often arise from axillary buds of leaves located at intermediate positions of extending shoots in many tree species including *Nothofagus* spp. (Remphrey & Powell 1984; Puntieri et al. 1998; Barthélémy et al. 1999; Costes & Guedon 2002; Marron et al. 2006; Costes et al. 2014; Puntieri et al. 2018) and may be considered an undesirable trait for timber production as they increases radically the number of knots on the trunk and, eventually, future timber defects (Ford 1985). However, sylleptic branches do not compete in height with their bearing axis, as they develop more horizontally and less vigourously than the one-year delayed (proleptic) branches; the latter are located in distal positions on trunk growth units and are more prone to be the origin of trunk forks (Barthélémy et al. 2007). Sylleptic and proleptic branches might compete for the available resources, which could be pre-empted by the former (Cline & Harrington 2007). This could explain the observed positive relationship between the hierarchical growth of the trunk and the development of sylleptic branches.

Plant height was also related to the mean internode length of the current trunk-GU, in agreement with the results of a previous study on *Nothofagus pumilio* (Magnin et al. 2017), which may suggest that internode length in trunk GU may contribute to the hierarchical architecture of a tree. On the other hand, our results show that the degree of hierarchical development assessed by the relative sizes reached by the current trunk-GU and a lateral-GU at the end of the growth season is not necessarily related to the extent of hierarchical architecture at the whole-tree level. It is possible that environmental factors modify the hierarchical structure of trees by mediating on traits such as the growth, sylleptic branching, apex persistence, and secondary growth of the trunk and the main branches of trees (Wu & Hinckley 2001).

Differences in the hierarchical growth of progenies derived from selected individuals

This work represents, to the best of our knowledge, the first attempt to evaluate variations in architectural traits of a *Nothofagus* species in progenies derived from controlled crosses of individuals that had been selected for their architecture. The fact that pollen viability as well as stigmatic receptivity is short in *Nothofagus* species (Torres & Puntieri 2013; García Cruzatty et al. 2015), together with the mentioned inter-annual variations in flower production (Torres et al. 2016), pose severe limitations to controlled crossings between selected trees. In this context, although our results cannot be extrapolated to wider scales, they could complement future studies in which higher numbers of open-pollination progenies could be assessed regarding their variations in architectural traits related to parent architecture. Plants that belonged to different progenies presented morphological variations in their GU after three growing seasons, although their basal diameters were similar. Some degree of variation in stem slenderness and mean internode length were related to pollen origin, ovule-source tree and to the time of extension of the GU. For instance, among the progeny of the *NH1* tree, stems tended to vary in slenderness and mean internode length depending on pollen origin in the case of GU2 but not in GU3. On the other hand, mean internode length for seedlings derived from the *H1* tree differed between pollen origins, but such variations were inconsistent for GU2 and GU3. This could be explained by speculating climatic variations between the growing seasons in which GU2 and GU3 extended and/or to the extent of acclimation of the study plants to outdoors conditions (considering that GU1 had developed in a glasshouse).

Some support to the idea that adult *N. obliqua* trees with hierarchical architectures could produce young trees showing incipient signs of a similar architecture may be found in the structure of GU3. This GU was slenderer and had longer internodes in the progeny derived from both hierarchical parent trees (*H1xH2*) than in the progeny of non-hierarchical parent trees (*NH1xNH2*). In accordance with the results obtained from forest saplings, stem slenderness and internode length would be positively linked with the hierarchical architecture of young individuals. These traits are relevant in terms of hydraulic conductance (Yoshimura 2011) and mechanical properties (Niklas 2007; Waghorn & Watt 2013) among other trunk functions (Yagi 2004; Puntieri & Ghirardi 2010). Even though internode length and slenderness could both vary in response to environmental pressures (Poorter & Rozendaal 2008; Torres et al. 2018), our results agree with previous studies, supporting the idea that a genetic component is involved in their determination (Costes & Gion 2015; Sun et al. 2019).

The indexes of hierarchical structure computed after three growing seasons ($H_{indexGU2}$ and $H_{indexGU3}$) did not vary consistently among progenies. Whereas $H_{indexGU3}$ tended to be higher in plants derived from the hierarchical ovule-donor tree (*H1*) irrespective of pollen origin, $H_{indexGU2}$ was similar among progenies and was only affected by basal trunk diameter. Therefore, our results suggest that the direct relationship between the hierarchical development of a parental tree and that of its progeny should not be discarded, although this was only evident, in this study, before axial secondary growth. Basal diameter at early ontogeny, which is often considered in the selection of vigourous individuals (Tsakaldimi et al. 2012), may not be a reliable trait for the selection of *N. obliqua* seedlings in cases where it is intended to generate trees with a hierarchical architecture (e.g. for timber production). In contrast, trunk diameter and hierarchical architecture are positively related in juvenile *N. obliqua* trees occurring in dense regeneration patches (this study), where competition for light would be a significant selective force. Whenever light is not a limiting factor, as in the nursery trees studied here, the largest plants could be variable in terms of hierarchical structure. Under such conditions, the combination of trunk vigour with low occurrence of codominant branches could be more suitable for seedling selection.

The frequency of seedlings with a very narrow angle between the main branch and the trunk (MBλ < 22.5°) was low in this survey, so that trunk forking was not likely for these plants. Unexpectedly, a higher frequency of wide insertion angles of the MB was found for the progeny derived from the ovule tree with a non-hierarchical architecture (*NH1*). The degree to which this could imply a lower susceptibility to trunk forking in the *NH1* progeny than in the *H1* progeny should be confirmed at more advanced ontogenic stages. Genetic variations of this trait have not been observed in a previous study performed on juvenile individuals of *N. nervosa,* a species closely related to *N. obliqua* (Puntieri et al. 2013a). Our current results, however, suggest that variations in the insertion angle of the main branch in *N. obliqua* seedlings could have a significant genetic component. This trend should be confirmed when the studied plants reach juvenile stages, given that this trait could present variations determined by ontogenetic factors (Puntieri et al. 2013b; Corvalán Vera 2017).

Sylleptic branching was frequent for GU with basal diameter >3 mm and variations were observed between plants derived from different pollen origins within the progeny of the *H1* tree. Overall, plants of the *H1 x H2* progeny developed more sylleptic branches than plants of the other progenies. Sylleptic branching in trees could be affected by genetic and environmental factors and may vary between closely-related species (Wu & Stettler 1998; Funnell et al. 2015) and even among varieties of the same species (Negrón et al. 2013). Our results agree with the hypothesis that sylleptic branching would be under genetic control, although environmental effects should also be considered. High production of sylleptic branches in trunk GU could negatively affect wood quality, but it also implies significant increments in leaf area during the growing season (Wu & Stettler 1998; Wu & Hinckley 2001), so that trunk differentiation relative to its main branches (i.e. a hierarchical architecture) would be promoted.

As a general trend, the progeny of the *H1* mother tree as well as the progeny derived from *NH1 x H2* crosses showed higher frequencies of shoot apex persistence in GU with low-diameter stems than the progenies derived from *NH1 x NH2* crosses and from open pollination of mother tree *NH1.* Therefore, this results agree with previous references suggesting endogenous factors related to apex death/persistence variations in *N obliqua* (Puntieri et al. 2007). Moreover, the fact that apex persistence was more frequent among plants derived from parent trees with a hierarchical structure (as suggested in the previous section), reinforces the idea that this variable could be a useful selection tool.

Conclusions

In juvenile individuals of *N. obliqua*, the hierarchical crown architecture was associated with morphological traits of the trunk growth units, such as the persistence of the apex between successive years, sylleptic branching and, indirectly, the average length of internodes. Both apex persistence and the production of sylleptic branches also presented variations related to parental origin in the case of seedlings. These traits seem to have a genetic basis and may be adopted as selection criteria at early ontogenetic stages for the development of *N. obliqua* trees with a hierarchical architecture. In seedlings, basal diameter of the trunk was negatively related to hierarchy, so on its own may not be as reliable a trait for the selection of seedlings of this species for timber production.

List of abbreviations

GU: Growth units
MB: Main branch

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

CT, JP and LG proposed the idea and experimental design. CT and JP selected trees for controlled crosses and performed manual pollinations. Seed processing, seedlings production and their subsequent transfer to the nursery were performed by TS. Plant measurements and data collection were performed by CT, AM, MStecconi, MSalgado, CS and JP. CT performed data analysis and wrote the initial draft of the manuscript which was revised by all co-authors.

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References

Aparicio, A.G., Zuki, S.M., Azpilicueta, M.M., Barbero, F.Á., & Pastorino, M.J. (2015). Genetic versus environmental contributions to variation in seedling resprouting in *Nothofagus obliqua*. *Tree Genetics Genomes, 11*(2): 23. https://doi.org/10.1007/s11295-015-0847-0

Azpilicueta, M.M., Marchelli, P., Aparicio, A.G., Pastorino, M.J., El Mujtar, V., Torres, C.D., Puntieri, J.G., Stecconi, M., Barbero, F., & Lozano, L. (2020). Roble pellín (*Nothofagus obliqua*): A southern beech with a restricted distribution area but a wide environmental range in Argentina. In M. Pastorino & P. Marchelli (Eds.), *Low Intensity Breeding of Native Forest Trees in Argentina: Genetic Basis for their Domestication and Conservation* (pp. 89-116). Switzerland: Springer. https://doi.org/10.1007/978-3-030-56462-9_4

Barthélémy, D., Puntieri, J., Brion, C., Raffaele, E., Marino, J., & Martinez, P. (1999). Características morfológicas y arquitecturales de las especies de *Nothofagus* Blume (Fagaceae) del norte de la Patagonia Argentina. *Boletín de la Sociedad Argentina de Botánica, 34*, 29-38.

Barthélémy, D., & Caraglio, Y. (2007). Plant architecture: a dynamic, multilevel and comprehensive approach to plant form, structure and ontogeny. *Annals of Botany, 99*(3), 375-407. https://doi.org/10.1093/aob/mcl260

Bates, D., Mächler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statical Software*, 67,1-48. https://doi.org/10.18637/jss.v067.i01

Cameron, A.D., Kennedy, S.G., & Lee, S. (2012). The potential to improve growth rate and quality traits of stem straightness and branching habit when breeding *Picea sitchensis* (Bong.) Carr. *Annals of Forest Science, 69*, 363-371. https://doi.org/10.1007/s13595-011-0167-y

Cline, M.G., & Harrington, C.A. (2007). Apical dominance and apical control in multiple flushing of temperate woody species. *Canadian Journal of Forest Research, 37*(1), 74-83. https://doi.org/10.1139/x06-218

Corvalán Vera, P. (2017). Caracterización del diámetro, ángulo de inserción y longevidad de ramas vivas axiales de *Nothofagus obliqua*. *Revista Cubana de Ciencias Forestales, 5*(2), 127-139.

Costes, E., & Guédon, Y. (2002). Modelling branching patterns on 1-year-old trunks of six apple cultivars. *Annals of Botany, 89*(5), 513-524. https://doi.org/10.1093/aob/mcf078

Costes, E., Crespel, L., Denoyes, B., Morel, P., Demene, M.N., Lauri, P.E., & Wenden, B. (2014). Bud structure, position and fate generate various branching patterns along shoots of closely related Rosaceae species: a review. *Frontiers in Plant Science, 5:* 666. https://doi.org/10.3389/fpls.2014.00666

Costes, E., & Gion, J.-M. (2015). Genetics and Genomics of Tree Architecture. *Advances in Botanical Research, 74*, 157-200. https://doi.org/10.1016/bs.abr.2015.05.001

Cumbie, W.P., Isik, F., & McKeand, S.E. (2012). Genetic Improvement of Sawtimber Potential in Loblolly Pine. *Forest Science, 58*(2), 168-177. https://doi.org/10.5849/forsci.09-060

Donoso, P., Donoso, C., Gallo, L., Azpilicueta, M., Baldini, A., & Escobar, B. (2006). *Nothofagus obliqua* (Mirb.) Oerst. Roble, Pellín, Hualle. In C. Donozo Zegers (Ed.), *Las especies arbóreas de los bosques templados de Chile y Argentina, autoecología* (pp. 471-485). Valdivia, Chile.

Drénou, C. (2000). Pruning trees: the problem of forks. *Journal of Arboriculture, 26*(5), 264-269. https://doi.org/10.48044/jauf.2000.032

Drénou, C., Restrepo, D., & Slater, D. (2020). Demystifying tree forks: vices and virtues of forks in arboriculture. *Journal of Botany Research, 3*(1), 100-113. https://doi.org/10.36959/771/566

Ford, E. (1985). Branching, crown structure and the control of timber production. In M. G. Cannell & J. E. Jackson (Eds.), *Attributes of trees as crop plants* (pp. 228-252). Abbotts Ripton: Intitute of Terrestial Ecology.

Funnell, K.A., Seleznyova, A.N., Kaji, R., Chen, J., Manandhar, S., & Woolley, D.J. (2015). Diverse growth and sylleptic branching patterns in Japanese maple cultivars. *Acta Horticulturae,* (1104), 143-148. https://doi.org/10.17660/ActaHortic.2015.1104.22

García Cruzatty, L., Rivero, M., & Droppelmann, F. (2015). Effect of temperature and drying on the longevity of stored *Nothofagus alpina* pollen. *New Zealand Journal of Botany, 53*(3), 155-164. https://doi.org/10.1080/0028825X.2015.1045528

Kenward, M., & Roger, H. (1997). Small sample inference for fixed effects from restricted maximum likelihood. *Biometrics*, *53*, 983-997 https://doi.org/10.2307/2533558

Magnin, A., Villalba, R., Torres, C.D., Stecconi, M., Passo, A., Sosa, C.M., & Puntieri, J.G. (2016). Effect of volcanic ash deposition on length and radial growths of a deciduous montane tree (*Nothofagus pumilio*). *Austral Ecology, 42*, 103-112. https://doi.org/10.1111/aec.12405

Magnin, A., Torres, C.D., Villalba, R., & Puntieri, J.G. (2017). Does water availability regulate biomass partitioning between trunk and branches? *Plant Biology*, *19*, (6), 917-925. https://doi.org/10.1111/plb.12614

Marchelli, P., Pastorino, M.J., & Gallo, L. (2020). Temperate Subantartic Forests: a huge natural laboratory. In M.J. Pastorino & P. Marchelli (Eds.), *Low Intensity Breeding of Native Forest Trees in Argentina - Genetic bases for their domestication and conservation* (pp. 27-54). Switzerland: Springer. https://doi.org/10.1007/978-3-030-56462-9_2

Marron, N., Bastien, C., Sabatti, M., Taylor, G., & Ceulemans, R. (2006). Plasticity of growth and sylleptic branchiness in two poplar families grown at three sites across Europe. *Tree Physiology, 26*(7), 935-946. https://doi.org/10.1093/treephys/26.7.935

Millet, J., Bouchard, A., & Édelin, C. (1999). Relationship between architecture and successional status of trees in the temperate deciduous forest. Écoscience, 6(2), 187-203. https://doi.org/10.1080/11956860.1999.11682520

Negrón, C., Contador, L., Lampinen, B.D., Metcalf, S.G., DeJong, T.M., Guédon, Y., & Costes, E. (2013). Systematic analysis of branching patterns of three almond cultivars with different tree architectures. *Journal of the American Society for Horticultural Science, 138*(6), 407-415. https://doi.org/10.21273/JASHS.138.6.407

Niklas, K. (2007). Maximum plant height and the biophysical factors that limit it. *Tree Physiology, 27*(3), 433-440. https://doi.org/10.1093/treephys/27.3.433

Osada, N. (2011). Height-dependent changes in shoot structure and tree allometry in relation to maximum height in four deciduous tree species. *Functional Ecology, 25*(4), 777-786. https://doi.org/10.1111/j.1365-2435.2011.01833.x

Poorter, L., & Rozendaal, D.M. (2008). Leaf size and leaf display of thirty-eight tropical tree species. *Oecologia, 158*(1), 35-46. https://doi.org/10.1007/s00442-008-1131-x

Puntieri, J., Barthélémy, D., Martinez, P., Raffaele, E., & Brion, C. (1998). Annual-shoot growth and branching patterns in *Nothofagus dombeyi* (Fagaceae). *Canadian Journal of Botany, 76*(4), 673-685. https://doi.org/10.1139/b98-041

Puntieri, J., Grosfeld, J.E., Stecconi, M., Brion, C., Azpilicueta, M.M., & Gallo, L. (2006). Desarrollo temprano del roble (*Nothofagus obliqua*): un análisis arquitectural de procedencias de Argentina. *Bosque, 27*(1), 44-51. https://doi.org/10.4067/S0717-92002006000100005

Puntieri, J., Grosfeld, J., Stecconi, M., Brion, C., Azpilicueta, M.M., Gallo, L., & Barthélémy, D. (2007). Shoot development and dieback in progenies of *Nothofagus obliqua*. *Annals of Forest Science, 64*(8), 839-844. https://doi.org/10.1051/forest:2007068

Puntieri, J., & Ghirardi, S. (2010). Growth-unit structure in trees: effects of branch category and position on *Nothofagus nervosa*, *N. obliqua* and their hybrids (Nothofagaceae). *Trees, 24*(4), 657-665. https://doi.org/10.1007/s00468-010-0435-8

Puntieri, J., Grosfeld, J., Tejera, L., Mondino, V., & Gallo, L. (2013a). Within-population variability in architectural traits and suitability to forestry conditions in *Nothofagus nervosa* (= *N. alpina*; Nothofagaceae). *Annals of Forest Science, 70*(5), 471-479. https://doi.org/10.1007/s13595-013-0286-8

Puntieri, J., Torres, C., & Ghirardi, S. (2013b). Crecimiento y ramificación de *Nothofagus alpina* y *Nothofagus obliqua* (Nothofagaceae) bajo diferentes condiciones lumínicas. *Boletín de la Sociedad Argentina de Botánica, 48*(2), 277-293. https://doi.org/10.31055/1851.2372.v48.n2.6262

Puntieri, J., Torres, C., Magnin, A., Stecconi, M., & Grosfeld, J. (2018). Structural differentiation among annual shoots as related to growth dynamics in *Luma apiculata* trees (Myrtaceae). *Flora, 249*, 86-96. https://doi.org/10.1016/j.flora.2018.10.005

R Development Core Team (2024). R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.

Remphrey, W., & Powell, G. (1984). Crown architecture of *Larix laricina* saplings: shoot preformation and neoformation and their relationships to shoot vigour. *Canadian Journal of Botany, 62*, 2181-2192. https://doi.org/10.1139/b84-298

Schinelli Casares, T. (2012). Producción de *Nothofagus* ajo condiciones controladas. Ediciones INTA, Esquel, 2012. 80 p.

Sola, G., Attis Beltrán, H., Chauchard, L., & Gallo, L. (2015). Efecto del manejo silvicultural sobre la regeneración de un bosque de *Nothofagus dombeyi*, *N. alpina* y *N. obliqua* en la Reserva Nacional Lanín (Argentina). *Bosque, 36*(1), 113-120. https://doi.org/10.4067/S0717-92002015000100012

Sola, G., El Mujtar, V., Tsuda, Y., Vendramin, G.G., & Gallo, L. (2016). The effect of silvicultural management on the genetic diversity of a mixed *Nothofagus* forest in Lanín Natural Reserve, Argentina. *Forest Ecology and Management, 363*, 11-20. https://doi.org/10.1016/j.foreco.2015.12.018

Soliani, C., & Aparicio, A.G. (2020). Evidence of genetic determination in the growth habit of *Nothofagus pumilio* (Poepp. & Endl.) Krasser at the extremes of an elevation gradient. *Scandinavian Journal of Forest Research, 35*(5-6), 211-220. https://doi.org/10.1080/02827581.2020.1789208

Sun, X., Shu, J., Ali Mohamed, A.M., Deng, X., Zhi, X., Bai, J., Cui, Y., Lu, X., Du, Y., Wang, X., Huang, Z., Guo, Y., Liu, L., & Li, J. (2019). Identification and characterization of EI (Elongated Internode) gene in tomato (*Solanum lycopersicum*). *International Journal of Molecular Sciences, 20*(9), 2204. https://doi.org/10.3390/ijms20092204

Torres, C., & Puntieri, J. (2013). Pollination and self-interference in *Nothofagus*. *Flora - Morphology, Distribution, Functional Ecology of Plants, 208*(7), 412-419. https://doi.org/10.1016/j.flora.2013.07.002

Torres, C., Magnin, A., Stecconi, M., & Puntieri, J. (2016). Testing individual inter-annual variations in flower production by means of retrospective analysis of meristem allocation in two tree species. *Folia Geobotanica, 51,* 361-371. https://doi.org/10.1007/s12224-016-9256-x

Torres, C., Magnin, A., Varela, S.A., Stecconi, M., Grosfeld, J.E., & Puntieri, J.G. (2018). Morpho-physiological responses of *Nothofagus obliqua* to light intensity and water status, with focus on primary growth dynamics. *Trees, 32*(5), 1301-1314. https://doi.org/10.1007/s00468-018-1712-1

Tsakaldimi, M., Ganatsas, P., & Jacobs, D.F. (2012). Prediction of planted seedling survival of five Mediterranean species based on initial seedling morphology. *New Forests, 44*(3), 327-339. https://doi.org/10.1007/s11056-012-9339-3

Vargas-Reeve, F., Mora, F., Perret, S., & Scapim, C.A. (2013). Heritability of stem straightness and genetic correlations in *Eucalyptus cladocalyx* in the semi-arid region of Chile. *Crop Breeding Applied Biotechnology, 13*, 107-112. https://doi.org/10.1590/S1984-70332013000200002

Waghorn, M.J., & Watt, M.S. (2013). Stand variation in *Pinus radiata* and its relationship with allometric scaling and critical buckling height. *Annals of Botany, 111*(4), 675-680. https://doi.org/10.1093/aob/mct015

Wu, R., & Stettler, R. (1998). Quantitative genetics of growth and development in *Populus*. III. Phenotypic plasticity of crown structure and function. *Heredity, 81*(3), 299-310. https://doi.org/10.1038/sj.hdy.6883970

Wu, R., & Hinckley, T. (2001). Phenotypic plasticity of sylleptic branching: genetic design of tree architecture. *Critical Reviews in Plant Sciences, 20*(5), 467-485. https://doi.org/10.1080/07352689.2001.10131827

Xiong, J.S., McKeand, S.E., Whetten, R.W., & Isik, F.T. (2014). Genetics of stem forking and ramicorn branches in a cloned loblolly pine family. *Forest Science, 60*(2), 360-366. https://doi.org/10.5849/forsci.12-018

Yagi, T. (2004). Within-tree variations in shoot differentiation patterns of 10 tall tree species in a Japanese cool-temperate forest. *Canadian Journal of Botany, 82*(2), 228-243. https://doi.org/10.1139/b03-124

Yoshimura, K. (2011). Hydraulic function contributes to the variation in shoot morphology within the crown in *Quercus crispula*. *Tree Physiology, 31*(7), 774-781. https://doi.org/10.1093/treephys/tpr060