

**RESEARCH ARTICLE** 

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# How much carbon do Argentine Pampas Pinus radiata plantations store?

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### Abstract

Aim of study: To quantify the biomass and carbon stored in the main ecosystem components in *Pinus radiata* D. Don plantations across an age sequence in the Pampean region of Argentina.

Area of study: Plantations were established on non-agricultural land, southeast of the province of Buenos Aires.

*Materials and methods:* Fourteen sites were selected of *Pinus radiata* plantations, 9-, 13-, 15-, 19- and 21-years-old, in a first forest rotation. Forty-two trees were destructively sampled, allometric functions were set and biomass was estimated for the different compartments (needles, branches, stem). Root biomass was estimated from equations adjusted by the sampling of twenty-four trees. At 4 sites, C-stock was determined in the tree component, in the forest floor and understory, and soil organic carbon (SOC) was determined to a 50 cm depth.

*Main results:* C-stock in the tree component increased with stand age, whereas SOC and C-stock in the forest floor and understory were not related to stand age. The system-level C-stock was 273.1, 263.7, 269.7 and 324.1 Mg ha<sup>-1</sup> for the 9-, 13-, 19- and 21-year-old stands. On average, 69% of the total system-level C-stock was in the soil, while 28% was in the tree biomass and 3% was in the forest floor and understory.

*Research highlights:* The forestry component contributed to C sequestration with no changes in SOC-stocks reserves for the age range studied.

Additional key words: forestry system; biomass accumulation; carbon sequestration; soil organic carbon; plantation age.

**Abbreviations used:** bb (branch biomass of the branch); BB (branches biomass of tree); BR (root biomass of tree); DBH (diameter at breast height); DM (dry matter); dr (diameter at the base of the branches); h (height); nb (needle biomass of the branch); NB (needle biomass of tree); SB (stem biomass);  $SOC_{meq}$  (soil organic carbon);  $SOC_{meq}$  (amount -or stock- of organic carbon in equivalent mass); RB (root biomass); v (individual volume); r (Pearson correlation coefficient).

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## Introduction

Earth air and ocean temperatures have increased because of the accumulation of gases in the earth's atmosphere. Indeed, there is straightforward evidence that carbon dioxide (CO<sub>2</sub>) concentrations are continuously rising; by 2019 they globally reached 410 ppm (IPCC, 2021). Thus, controlling the amount of CO<sub>2</sub> has become crucial. In this direction, two strategies can be considered: on the one hand, reducing or avoiding greenhouse gas (GG) emissions, and on the other hand, increasing carbon (C) sequestration (Olmedo et al., 2020).

Given that forests function as C reservoirs (Dixon et al., 1994), practices such as afforestation and reforestation are essential. Forests perform multiple simultaneous functions such as producing high quality timber, fuelwood, non-timber forest products and fiber. They offer jobs, clean water, and air; buffering noises, drought and floods; they also provide the habitat for both human recreation and wildlife biodiversity (Benz et al., 2020).

In forest ecosystems, C accumulation depends on the interactions between the stand structure and the site quality. To adjust forest management strategies for maintaining or increasing C stocks, or making projections, it is crucial to understand the magnitude of C sequestration by forest systems of different development stages under different soil and climatic conditions (Merino et al., 2003).

One of the C reservoirs in forest ecosystems is their biomass. To accurately assess C at the system level requires quantifying the C in each component. However, the tree component C estimation is a laborious and costly process if done directly. In the case of forest systems, the most frequent way to estimate the biomass in the tree (needles, branches, roots, and stem) is using equations taken from destructive sampling of individuals at different ages and dimensions (Balboa-Murias et al., 2006). Though, to get a complete picture of the system C-stocks, quantifying components such as soil and the forest floor is also necessary (Zhao et al., 2014; Smal et al., 2019; Zhang et al., 2019).

Indeed, soil is an important C reservoir. According to Dixon et al. (1994), on average, 69% of C-stock is stored as soil organic matter and 31% is accumulated as living biomass. At the site level, factors like soil texture control, soil organic carbon (SOC) stocks (Zhou et al., 2019). However, other factors such as plantation age and forest management can modify the amount of C-stock accumulated. Regarding age, Paul et al. (2002) reported that SOC decreases in the first five years after afforestation, followed by a recovery period to reach its peak among their 20-50 years, depending on the environmental conditions. However, in contrast to the SOC labile fractions, the more stable fractions, on which the C-stock inventories are based, do not express short-term variations. For this reason, it is important to analyze chronosequences that may model what would happen in a rotation (Zhao et al., 2014; Smal et al., 2019; Zhang et al., 2019).

Forest floor and understory are also key components because of their functions in the forest ecosystem; in addition to regulating the levels of SOC, they are the source of energy for soil microflora and fauna and control the release of nutrients and their availability to plants (Vesterdal et al., 2002).

The study of C-stock accumulation patterns during the development of a forest system has become increasingly important, particularly in those environments where forest plantations replaced other land uses. In the Pampas region of Argentina, grasslands are the natural vegetation (Soriano, 1991). They are used as a base for livestock, or replaced by pastures, also for livestock purposes or for agriculture. In the mountain areas, where soils are shallow, some area of grasslands have been replaced by pine plantations (especially Pinus radiata in the Tandilia system, and Pinus halepensis in the Ventania system) to timber production and to contribute to the development of the local industry. However, few studies have examined the contribution of these forest systems to C-stock sequestration. In a wide geographical region, Zalba & Peineman (1987) and Berhongaray et al. (2013) compared C-stocks across different land uses. Álvarez et al. (2021) suggested that plantation soils may still have C sequestration potential when these plantations are established. The objectives of this study were to: 1) provide equations to estimate different biomass component (needles, branches, stem and roots) of P. radiata trees, and 2) determine the C stored in the main components of P. radiata plantations across an age sequence.

## **Material and methods**

### Study area

This study was conducted in the SE of the Buenos Aires province, Argentina (37°18′05.05"S 38°44′39.14"S) (Fig. 1), a highly productive region. The region belongs to the Pampa and field's ecoregion, where the dominant original vegetation is *Piptochaetium montevidense, Stipa neesiana, Panicum* spp., *Bothriochla laguroides* and *Poa* spp. (Apodaca et al., 2015).

In the province of Buenos Aires, most of the commercial plantations of *P. radiata* are located in the 'Sierras de Tandil' chain and, to a lesser extent in 'Sierras de Ventania'. The climate is temperate, with year-round rainfalls (885 mm), average temperatures below 22 °C in the warmest month (Köppen-Geiger). The average annual temperature is 13.9 °C.

For the study, *P. radiata* in the first forest rotation stands 9, 13, 15, 19, and 21 year old were identified in the 'Sierras de Tandil' (Fig. 1). In general, stands presented high initial density and unmanaged forest (no pruning and no thinning). Sample sites and descriptive features of the actual state of



Figure 1. Location of Pinus radiata plantations in the study area.

each stand are detailed in Table 1. In some stands with lower densities (e.g., sites 13 and 14), a higher mortality due to competition was observed, reaching current densities that are detailed in Table 1. There were three replicates for each age, except for the 19-year-old stand where there were two replicates. There were 14 sampling sites located in mountain areas of less than 500 m a.s.l. altitude and slopes of 5-10%. The distance between the sampling site was 5 to 30 km and the area of the smallest stand was 5 ha. The soil where all plantations were located is a *Petrocalcic argiudol* (Soil Survey Staff, 2014), with important SOC levels, superficial clay loam texture, with effective depth of 30-60 cm, due to the presence of coarse and rocky outcrops, well drained, formed on loess sediments.

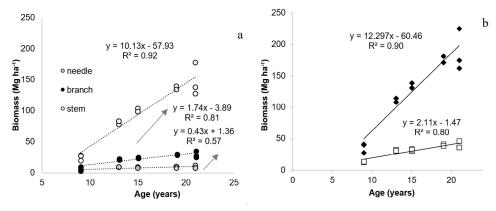
#### Forest inventory and measurements

In each sampling site we established between three and six inventory plots (depending on the size of the stand) of 400 m<sup>2</sup> (20 m × 20 m). Within each plot, we measured the diameter at breast height (DBH) of all trees and the height (h) of the four thickest trees, one medium DBH tree and one lower DBH tree. H is the dominant height and was calculated as the average of the 100 thickest trees (the average of four thickest trees of the plot). With DBH-h (n=219) from the different sites and ages studied, a model was developed to estimate the height of individual trees. The model is: ln y =  $a + b^* \ln (x_1) + c^* \ln(x_2)$ , where y is height (h, m), a, b and c are equation parameters, x<sub>1</sub> is DBH (cm),

Table 1. Sites and descriptive features of the Pinus radiata stands.

Stand	Age (years)	D (tree ha <sup>-1</sup> )	H (m)	d <sub>g</sub> (cm)	v (m <sup>3</sup> ha <sup>-1</sup> )
1	9	$606\pm26$	$10.0\pm0.4$	$20.7\pm0.5$	$79.1\pm6.0$
2	9	$783\pm77$	$9.2\pm0.3$	$17.2\pm0.6$	$78.9 \pm 16.7$
3	9	$561\pm38$	$9.2\pm0.2$	$21.1\pm0.4$	$76.7\pm5.6$
4	13	$1450\pm 66$	$12.5\pm0.1$	$20.0\pm0.1$	$220.2\pm8.1$
5	13	$1366\pm68$	$13.4\pm0.3$	$21\pm0.4$	$235.2\pm20.6$
6	13	$1325\pm22$	$12.1\pm0.5$	$21.2\pm0.1$	$234.4\pm5.3$
7	15	$1176\pm104$	$14.1\pm0.1$	$22.2\pm0.3$	$252.8\pm15.6$
8	15	$1166\pm72$	$14.1\pm0.2$	$23.4\pm0.6$	$293.4\pm9.6$
9	15	$1056\pm122$	$16.2\pm0.2$	$24.0\pm0.5$	$279.8\pm 21.0$
10	19	$1360\pm46$	$16.1\pm0.2$	$23.7\pm0.3$	$394.6\pm10.7$
11	19	$1360\pm122$	$16.2\pm0.7$	$22.8\pm1.3$	$396.4\pm20.0$
12	21	$1137\pm51$	$18.9\pm0.3$	$27.5\pm 0.8$	$503.9\pm14.8$
13	21	$870\pm25$	$18.6\pm0.6$	$27.7\pm 0.3$	$394.4\pm5.3$
14	21	$870\pm35$	$18.2\pm0.2$	$27.5\pm 0.3$	$390.5\pm15.6$

D: actual density. H: dominant height. dg: quadratic mean diameter. v: volume. The value that follows  $\pm$  is the standard error.



**Figure 2.** Relationship between: (a) stand age and biomass of needles, branches, and stems; b) stand age and root biomass, aboveground biomass.

and  $x_2$  is the tree age (years). With this equation, the h of all the trees in the plots was estimated and used to predict the volume of each tree in the plot.

### **Estimation of tree biomass**

To estimate needle biomass (NB) and branch biomass (BB), 42 trees were destructively sampled from the sites indicated in Table 1. Three radiata pine trees representing the range in DBH and height distribution in each stand were selected for felling. In each tree, the diameter at the base of all branches (dr) was measured. One branch per whorl was randomly selected, needle and branch biomass were determined separately in situ, and samples of each component  $(\approx 300 \text{ g})$  were collected and oven-dried at 65° C to constant weight for moisture content. The models used to estimate the dry matter of needles (nb) in each branch and branches (bb), as a function of the diameter of each branch (dr), was  $\ln y = a + b^* (\ln dr)$ , where y is the dry matter (g) of the tree component (branches and needles), a and b are the equation parameters. Similar models were used to estimate the biomass fractions of needles (NB) and branches (BB) of each tree using the DBH.

To estimate the individual volume (v, m<sup>3</sup>) of each tree in the plot we used the following equation:  $v=a+b^*(DBH^{2*}h)$ , *a* and *b* are the equation parameters. To develop this model, 146 trees were felled and the main stem was cut into 1-m sections. The volume with bark of each section was calculated using the Smalian formula (De León & Uranga-Valencia, 2013). The volumes of each tree in the plot were estimated and added. With the surface of the plot, the value per hectare was calculated. Stem biomass (SB) was calculated by multiplying the stem volume by the average stem wood density. Wood density (kg m<sup>-3</sup>) was obtained from a random sampling of 21 trees from site 14. The sample of wood density tree was taken at DBH with Pressler auger. The mean density of trees was of  $353 \pm 56$  kg m<sup>-3</sup>.

The model used to estimate the root biomass (RB) in each tree was  $\ln y = a + b^* (\ln x)$ , y is the root biomass (kg), *a* and *b* are the equation parameters, *x* is DBH (cm) of each tree. To develop this model, 22 trees with DBH ranging from 15 to 45 cm, distributed in 3 diameter classes (11 trees in the 15-25 cm class; 6 trees in 26-35 cm class and 5 in 36-45 cm class) were sampled at site 14. An excavator was used for the extraction, which was completed with pick and shovel by digging up all roots larger than 1-2 cm in diameter. Roots were cleaned, weighed on a field scale (precision to the kilogram) and a sample was set aside to determine the dry matter (DM), at 65 °C to constant weight. The root biomass of each tree in the plot was estimated and added. With the surface of the plot, the value per hectare was calculated.

Total biomass at stand level was calculated as the addition of aboveground biomass (needles + branches + stem), and the root biomass. The amount of C was estimated assuming that 50% of the biomass is C (IPCC, 2003).

#### Biomass in forest floor and understory

Sampling for forest floor and understory was conducted at sites 3, 4, 11 and 14, corresponding to ages 9, 13, 19 and 21 years old. At site 7, 15-year-old sampling was not conducted as the forest floor had been periodically extracted for use in nursery and composting (*personal communication* from the owner). Forest floor and understory were sampled through collecting the material within four 1 m × 2 m subplots randomly chosen in each of three inventory plots (12 samples in each site). The material was weighed in situ and a subsample of each component was placed in an oven at 65°C until constant weight to determine the moisture content. DM values were extrapolated to the hectare (Mg ha<sup>-1</sup>). The amount of C was estimated assuming that 50% of the biomass is C (IPCC, 2003).

#### **Mineral soil sampling**

Soil sampling for SOC quantification was conducted at sites 3, 4, 11 and 14, corresponding to ages 9-, 13-, 19- and

-0.1841			$\mathbf{R}^{2}_{adj}$	RMSE	$\overline{E}$	$ ar{E} $	p(a)	p(b)	p(c)
-0.1641	0.35416	0.60322	0.95	0.09	0.03	0.85	< 0.001	< 0.001	< 0.001
2.72618	2.844436		0.75	0.26	0.06	0.31	< 0.001	< 0.001	
3.15001	1.83651		0.91	0.15	0.11	0.49	< 0.001	< 0.001	
4.1151	1.54295		0.59	0.19	6.54	56.59	< 0.001	< 0.001	
2.36456	2.41072		0.68	0.32	36.59	191.30	< 0.001	< 0.001	
-3.33785	2.12634		0.88	0.05	0.01	0.35	< 0.001	< 0.001	
-0.0093	0.00003521		0.99	0.04	1.04	0.02	< 0.001	< 0.001	
	3.15001 4.1151 2.36456 -3.33785	3.150011.836514.11511.542952.364562.41072-3.337852.12634	3.15001       1.83651         4.1151       1.54295         2.36456       2.41072         -3.33785       2.12634	3.150011.836510.914.11511.542950.592.364562.410720.68-3.337852.126340.88	3.150011.836510.910.154.11511.542950.590.192.364562.410720.680.32-3.337852.126340.880.05	3.150011.836510.910.150.114.11511.542950.590.196.542.364562.410720.680.3236.59-3.337852.126340.880.050.01	3.150011.836510.910.150.110.494.11511.542950.590.196.5456.592.364562.410720.680.3236.59191.30-3.337852.126340.880.050.010.35	3.15001 $1.83651$ $0.91$ $0.15$ $0.11$ $0.49$ $<0.001$ $4.1151$ $1.54295$ $0.59$ $0.19$ $6.54$ $56.59$ $<0.001$ $2.36456$ $2.41072$ $0.68$ $0.32$ $36.59$ $191.30$ $<0.001$ $-3.33785$ $2.12634$ $0.88$ $0.05$ $0.01$ $0.35$ $<0.001$	3.15001       1.83651       0.91       0.15       0.11       0.49       <0.001

Table 2. Allometrics equations parameters to estimate needles, branch and root biomass, height and volume.

<sup>[1]</sup> *Y*: estimated variable, h: height (m); nb: needles biomass per branch (g branch<sup>-1</sup>); bb: branches biomass per branch (g branch<sup>-1</sup>); NB: needle biomass of tree (kg tree<sup>-1</sup>); BB: branches biomass per tree (kg tree<sup>-1</sup>); RB: root biomass (kg tree<sup>-1</sup>); *v*: volume (m<sup>3</sup> tree<sup>-1</sup>). R<sup>2</sup><sub>adj</sub>, coefficient of determination adjusted. RMSE, root mean squared error.  $\overline{E}$ :bias.  $|\overline{E}|$  mean of the absolute values of the residuals. p(a), p(b), p(c): p-value of parameters a, b, c. Equations are expressed as: ln h = *a*+*b*\**ln* (DBH) +*c*\*ln(age); ln lb (or bb) = *a*+*b*\*(ln dr); ln NB (or BB or RB) = *a*+*b*\*(ln DBH); v = *a*+*b*\*(DBH<sup>2</sup>h).

21-years old. In each inventory plot, a composite sample was taken at the depths of 0-10 cm, 10-25 cm, and 25-50 cm. Organic carbon (OC) concentration was determined using the dry combustion method (LECO Corporation, St Joseph, MI, USA). For these depths, bulk density was determined (Burke et al., 1986). The SOC mass in each soil stratum was calculated with the following formula:

$$OCM = BD * z * OC * 10$$

where OCM: OC mass content (Mg ha<sup>-1</sup>); BD: bulk density (Mg m<sup>-3</sup>); z = thickness of the layer sampled (m); and OC: organic carbon concentration (g kg<sup>-1</sup>).

To quantify SOC stocks at the system level, a mass correction was performed to bring soil profiles to equivalent mass ( $SOC_{meq}$ ) at the depth of 50 cm (Sisti et al., 2004).

#### Statistical analysis

The goodness of fit indicators used for model selection were  $R^2_{adj}$ ; root mean squared error (RMSE), bias ( $\overline{E}$ ) and mean of the absolute values of the residuals (MAR):

MAR: 
$$|\overline{E}| = \left|\frac{\sum_{i=1}^{N} E_i}{N}\right|$$
  
Bias:  $\overline{E} = \frac{\sum_{i=1}^{N} E_i}{N}$ 

Due to the focus of this study of analyzing allometric relationship between tree biomass components across stand age, additivity of the biomass equations was not considered (Yang et al., 2019).

One-way analysis of variance (ANOVA) followed by the Tukey's test at the 0.05 significance level was used to detect the mean difference to biomass component, concentration and SOC stock in each layer, the  $SOC_{meq}$  stock at the 0-50 cm layer, forest floor and understory and carbon stock system component, among different stand age. Previously, normality by Shapiro-Wilks and homogeneity of variance by Levene Test were verified.

Pearson correlation analysis (r) was applied to determine the relationships to biomass in the different tree components (needles, branches, stems and roots), aboveground biomass, total biomass, C content in different system components (SOC<sub>meq</sub> stock, C in the tree component, C in the forest floor), and the age of the plantations. When the correlation was significant, simple regression models were established to analyze the relationship between each variable and age. Statistical analyses were performed with Infostat software (Di Rienzo et al., 2008).

## Results

#### **Tree models**

The parameters of the equations developed to estimate the DM of needles and branch as a function of dr were calculated (Table 2), and they were significant to 99.9% level (p<0.001); a and b coefficients also resulted equally significant (p<0.001). Based on the data obtained from equations nb and bb, the biomass for each component (needles and branches) of all the branches measured was estimated; their sum constitutes the total biomass of needles and branches of each tree. The equations NB, BB and RB were used to determine the biomass of each tree component (needles, branches, and roots). The allometric equations were developed satisfactorily and explained the variability in root, branch and to a lesser extent in needle components. Using the logarithmic regression equations, the needle, branch and root biomass, height and volume was estimated for each tree of all radiata pine stands plot. By including DBH and age in the model, 95% of the varа

d

a

21

cd

а

19

**Figure 3.** Biomass percentage distribution of the different components in 9-, 13-, 15-, 19- and 21-year-old *Pinus radiata* stands. Different letters reflect statistically significant differences (p < 0.05) across different age stands.

■Root ■Stem □Branch ■Needle

с

ab

b

ab

13

b

9

h

ab

c

15

Stand age (years)

iability in tree height can be explained. For volume, the precision is higher. With the DBH and h variables involved in the model, the variability in the volume of the tree can be predicted by 99%.

#### **Biomass in the different tree components**

In the 9-year-old stands the total stand biomass, aboveground biomass and root biomass were 49.5, 36.5 and 12.9 Mg ha<sup>-1</sup>, respectively (Table 3). In the 21-year-old stands the total stand biomass, aboveground biomass and root biomass were 226.8, 187.2 and 39.6 Mg ha<sup>-1</sup>. Stem biomass was 24.8, 81.2, 100.4, 136.6 and 147.9 Mg ha<sup>-1</sup> in the 9-, 13-, 15-, 19-, and 21-year-old stands. Needle biomass was 3.5, 9.0, 8.7, 9.6 and 9.3 Mg ha<sup>-1</sup> for 9-, 13-, 15-, 19-, and 21-year-old stands. Biomass in branches for the range of ages studied was 8.2, 22.3, 24.1, 29.9 and 29.9 Mg ha<sup>-1</sup>, respectively.

A strong positive (p<0.05) and direct correlation (r) was found between the stand age and the needle bio-

mass (r=0.75), the branch biomass (r=0.90) and the stem biomass (r=0.96). Regressions for all radiate pine tree components (needle, branch and stem) were highly significant (p<0.001), with R<sup>2</sup> ranging from 0.5 to 0.9 (Fig. 2a). Similar results were obtained when considering the stand age relationship with RB (r=0.95) and aboveground (r=0.95). Regressions for this tree components (RB and aboveground biomass) with age, were highly significant (p<0.001), with R<sup>2</sup> between 0.8 to 0.9 (Fig. 2b).

The biomass allocation pattern follows the order: needle<branch<root<stem (Fig. 3). The biomass proportion of each component varied across the ages analyzed. In the needles, root, and branch components the proportion of biomass decreased significantly (p<0.05) with the age of the plantation. Between the ages of 9- and 21-years old, the proportion of biomass in needles dropped from 7.1% to 4.1%; in the roots, it changed from 27.2% to 17.5%, while the proportion of biomass in branches decreased from 16.5% to 13.2%. In contrast, the percentage of stem biomass increased significantly (p<0.05) from 49.0% to 65.1% in the 9- and 21-year-old stands, respectively.

The ratio of crown biomass (needles + branch biomass) to aboveground biomass decreased with stand age, being 32.5%, 27.8%, 24.6%, 22.4% and 20.9% at 9-, 13-, 15-, 19- and 21-year-old, respectively. The relationship of this crown ratio and v was significant (r=-0.99; p<0.05). A lineal regression for these variables were highly significant (p<0.001), with R<sup>2</sup> = 0.98. The ratio of root biomass to stem biomass decreased with stand age, being 53.1%, 38.6%, 31.9%, 29.1% and 26.8% at 9-, 13-, 15-, 19- and 21-year-old, respectively. The ratio of needles + branch biomass to stem biomass decreased slightly with stand age, being 48.7%, 48.8%, 48.2%, 47.5% and 46.8% at 9-, 13-, 15-, 19- and 21-year-old, respectively.

#### Carbon concentration and storage in the soil layer

SOC concentration decreased with profile depth (Table 4). For all depths, the lowest SOC concentrations (p<0.05) were found at the 19-year-old stands. No significant dif-

**Table 3.** Estimated biomass (Mg ha<sup>-1</sup>) of the tree components in the 9-, 13-, 15-, 19-, and 21-year-old radiata pine (*Pinus radiata* L.) stands.

Component	Stand age (years)						
	9	13	15	19	21		
Aboveground	$36.6 \pm 4.4$ a	$112.5 \pm 2.2 \text{ b}$	$133.2 \pm 2.3 \text{ bc}$	$171.1\pm4.9\ cd$	$187.2 \pm 19.4 \text{ d}$		
Needles	$3.5\pm0.3$ a	$9.0\pm0.1\;b$	$8.7\pm0.2\ b$	$9.6\pm1.0\;b$	$9.3\pm1.0\ b$		
Branch	$8.2 \pm 1.3$ a	$22.3\pm0.4\ b$	$24.0\pm0.4\ b$	$29.9\pm1.1\;b$	$29.9\pm3.1\ b$		
Stem	$24.8\pm2.7$ a	$81.2\pm1.7~\mathrm{b}$	$100.4\pm1.7~bc$	$136.5\pm2.7~\text{cd}$	$147.9\pm5.3~d$		
Root biomass	$12.9\pm0.5~a$	$31.4\pm0.4b$	$32.1\pm0.9\;b$	$39.8\pm1.4\ b$	$39.6\pm3.4\ b$		
Total	$49.5\pm4.9~a$	$143.9\pm2.6\ b$	$165.3\pm3.1\ bc$	$215.9\pm6.3\ cd$	$226.8\pm22.8\ d$		

Mean values with the standard error followed by different uppercase letters are significantly different at p < 0.05.

6

§ 100%

80%

60%

40%

20%

0%

Tree component biomass/Total biomass ×

9	13	19	21
SOC conc	entration (g kg <sup>-1</sup> )		
$63.5\pm2.5~b$	$60.5\pm3.7~b$	$41.8\pm3.7a$	$64.8\pm4.8\;b$
$49.5\pm3.0\ b$	$41.1\pm2.7\ ab$	$32.8\pm1.6\;a$	$47.2\pm3.0\ b$
$35.9\pm3.7\ b$	$23.0\pm2.5~a$	$22.2\pm2.5~a$	$26.3 \pm 2.8$ ab
SOC st	ock (Mg ha <sup>-1</sup> )		
$64.3\pm3.4\ b$	$53.6\pm4.7\ ab$	$43.9\pm3.6\;a$	$63.6\pm6.1b$
$87.1\pm4.3\ b$	66.1 ±6.8 ab	$53.0\pm4.4\ a$	$80.9\pm\!\!5.6~b$
$120.7\pm12.1~\mathrm{b}$	$69.3 \pm 7.2$ a	$58.2\pm8.8$ a	$78.0\pm9.3~a$
	<b>SOC conc</b> $63.5 \pm 2.5$ b $49.5 \pm 3.0$ b $35.9 \pm 3.7$ b <b>SOC st</b> $64.3 \pm 3.4$ b $87.1 \pm 4.3$ b	SOC concentration (g kg <sup>-1</sup> ) $63.5 \pm 2.5$ b $60.5 \pm 3.7$ b $49.5 \pm 3.0$ b $41.1 \pm 2.7$ ab $35.9 \pm 3.7$ b $23.0 \pm 2.5$ aSOC stock (Mg ha <sup>-1</sup> ) $64.3 \pm 3.4$ b $53.6 \pm 4.7$ ab $87.1 \pm 4.3$ b $66.1 \pm 6.8$ ab	SOC concentration (g kg <sup>-1</sup> ) $63.5 \pm 2.5$ b $60.5 \pm 3.7$ b $41.8 \pm 3.7a$ $49.5 \pm 3.0$ b $41.1 \pm 2.7$ ab $32.8 \pm 1.6$ a $35.9 \pm 3.7$ b $23.0 \pm 2.5$ a $22.2 \pm 2.5$ a           SOC stock (Mg ha <sup>-1</sup> ) $64.3 \pm 3.4$ b $53.6 \pm 4.7$ ab $43.9 \pm 3.6$ a $87.1 \pm 4.3$ b $66.1 \pm 6.8$ ab $53.0 \pm 4.4$ a

**Table 4.** Concentration and soil organic carbon stock in 0-10, 10-25 and 25-50 cm depths, at 9-, 13-, 19-, and 21-year-old *P. radiata* stands (mean  $\pm$  standard error).

Different letters reflect statistically significant differences ( $p \le 0.05$ ) across different age stands for the same profile depth.

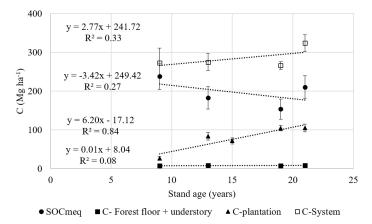
ferences (p>0.05) in SOC concentration at 0-10 and 10-25 were found cm between the 9-, 13- and 21-year-old stands. At 25-50 cm the highest concentration was found at 9 year-old stand and the lowest concentration at 13- and 19-year-old stand. When expressed in mass, the trend was similar to the observed when the values were expressed in concentration. No significant differences (p>0.05) were found in SOC stock in the 0-10 and 10-25 cm layer for the 9-, 13- and 21-year-old stands. The SOC stock stored at 0-25 cm represented between 56 and 65 % of the SOC stock at 0-50 cm.

#### Carbon in the system

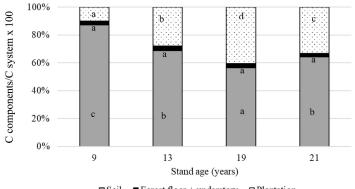
System-level C was defined as the sum of C tree components plus C forest floor and understory and  $SOC_{meq}$ stock in the 0-50 cm soil layer. The analysis at the system level was carried out considering the stands of sites 3, 4, 11 y 14 with ages of 9, 13, 19 and 21 years (Table 5). The 15-year-old stand at site 7 was not included because information on the amount of C in the forest floor and understory component was not available. The C-stock of plantation was 26.71, 72.2, 107.9 and 105.19 Mg ha<sup>-1</sup> and was directly and positively associated with the age (r=0.89; p<0.05). The model considering age explained 84% of the variation (R<sup>2</sup>=0.84, Fig. 4) of carbon stock in the plantation. C-stock in the forest floor and understory was 8.0, 8.4, 8.1 and 8.3 Mg ha<sup>-1</sup> for ages 9-, 13-, 19- and 21-year-old, respectively. The model including age only explained 8% of the variation (R<sup>2</sup>=0.08; Fig. 4) of carbon stock in this component. SOC<sub>meq</sub> stock at 0-50 cm depth was not associated with plantation age (r=-0.30; p>0.05).

The total system C stock was 273.1, 263.7, 269.7 and 324.1 Mg C ha<sup>-1</sup> for ages 9, 13, 19, and 21-year-old, respectively. The total system C stock showed no relationship with plantation age (r=0.40; p>0.05).

The C stock fractions across the stand were in the following order: soil > tree > forest floor and understory. The proportion of SOC<sub>meq</sub> in the soil to the total C-stock system were 88%, 69%, 56%, 64% for stands of 9, 13, 19 and 21 years, respectively (Fig. 5) and decreased significantly (p<0.05) up to 19 years old. The proportion of C in the tree component to the total system increased significantly



**Figure 4.** Relationship between the different system components C stock and the plantation age.



■ Soil ■ Forest floor + understory □ Plantation

**Figure 5.** Carbon proportion in the system components (plantation, forest floor and understory, and soil) at 9-, 13-, 19- and 21-year-old *Pinus radiata* plantations. Different letters reflect statistically significant differences (p < 0.05) across different age stands.

(p<0.05) up to 19- years old and the value was 10%, 28%, 40% and 33% for the 9-, 13-, 19- and 21-year-old stands, respectively (Fig. 5). Forest floor and understory as a proportion of total system C-stock ranged from 2.6% to 3.2% and did not show significant change with age (p>0.05).

# Discussion

### **Tree models**

Models that estimate the biomass in different components, h and volume include the DBH. In general terms, the models that use only the diameter as an independent variable present higher relative errors; while the incorporation of height (such as volume) significantly improves the prediction. For total volume with bark, the combination of the quadratic diameter with height both present the best results. For the purposes of accuracy, precision and ease of interpretation, the model selected is an expression of the cylinder affected by a shape factor. The correlation was higher (R<sup>2</sup>≥0.88) for the equations that estimate h, bb, RB and v (p<0.0001); and was lower (R<sup>2</sup>=0.59-0.75) for the equations that estimate nb, NB and BB (p<0.0001). Baker et al. (1984) adjusted equations that were considered suitable for their use in regional surveys: biomass of needles, total branches, stem bark, and total stem were given as functions of DBH, diameter at the base of the live crown, and total height. Stem biomass presented high R<sup>2</sup> with the DBH or with the DBH<sup>2</sup>h. Moore (2010) fitted aboveground biomass equations for *P. radiata* and showed the existence of strong relationships to DBH<sup>2</sup>h.

In our study, h or age were not included in NB, BB and RB, but its inclusion probably would have improved the estimation. Previous studies with other pines also reported  $R^2$  values over 95% calculated using the same equation for needles biomass, stem biomass, root biomass and branch biomass of Korean pine plantations in the central region of Korea (Li et al., 2011), Zhao et al. (2014) found that DBH explain more than 90% of the variability of all biomass components in young and mature stands of *Pinus tabulae-formis*.

#### **Biomass in the different tree compartments**

Stand age is the main factor affecting biomass magnitude and distribution in the forest system (Peichl & Arain, 2006; Lee et al., 2016). In our study, biomass of all tree component correlates positively and significantly with stand age. This is consistent with reports by Lee et al. (2016) and Zhang et al. (2019), and proves that age is an effective variable to accurately estimate biomass in the dif-

	Stand age (years)					
	9	13	19	21		
Tree C	$26.7\pm4.7~a$	$72.2\pm9.9~b$	$107.9\pm7.0~\mathrm{c}$	$105.2\pm9.9~\text{c}$		
Forest floor and understory C	$8.0\pm2.9~a$	$8.4 \pm 1.1$ a	$8.1\pm1.76$ a	$8.3\pm0.53\;a$		
$SOC_{meq} 0-50 \text{ cm}$	$238.4\pm13.7~\text{c}$	$182.9\pm11.8~ab$	$153.7 \pm 3.6 \text{ a}$	$210.6\pm12.2\ bc$		
Total C	$273.1\pm13.8 \text{ ab}$	$263.7 \pm 14.3 \text{ a}$	$269.7\pm4.3\ ab$	$324.1{\pm}~14.9~b$		

Different letters reflect statistically significant differences (p < 0.05) across different age stands.

ferent tree components. At the 21-year-old stand, the total biomass at the stand level was 4.5 times greater than the biomass at the 9-year-old one, while the aerial biomass and root biomass were 5 and 3 times higher, respectively. The biomass of the 19-year-old stands (Table 2) was slightly higher than that reported by Merino et al. (2003). These authors reported aerial biomass values of 149 Mg ha<sup>-1</sup> for a 19-year-old stand on a 50-cm effective depth medium-textured soil. Compared to other species of the genus Pinus, we can observe that P. radiata plantations accumulate a significant amount of biomass in short rotations. Biomass values reported in this study were higher than those reported by Zhang et al. (2019) for a 19-year in Pinus sylvestris L. plantations on sandy textured soils. They were also higher than those reported by Li et al. (2011) for 19-yearold stand of Pinus koraiensis on sandy loam-textured soils. The high productivity in terms of aerial biomass imply a great demand for water and nutrients and their supply is site dependent. Thus, the higher biomass in our plantations may be the result of their best site quality, with high organic matter content and higher supplier nutrient soils in a warmer climate (Schlatter & Gerding, 1999; Romanya & Vallejo, 2004). Laclau (2003a) found for a range of 10- and 20-year-old Pinus ponderosa plantations, that the stand biomass was between 8.3 and 144.5 Mg ha<sup>-1</sup>, respectively.

The patterns of biomass distribution among various components of the tree were in an order of stem wood> roots>branch> needles. This pattern is frequently reported in the literature. Similar research results were observed in an age sequence of P. tabulaeformis stand (Zhao et al., 2014) and P. sylvestris stand (Zhang et al., 2019). Laclau (2003a) reported in the same line for ponderosa pine plantations of 10 and 20-year-old respectively. In addition, this author indicated that the stem was the largest component of the total biomass (50%-65%), while the crown (branches+needles) varied from 25% to 17%. These percentages are similar to those reported in our study for stands of 9 and 21 years old, where the proportion of stem was between 49.0% and 65.1% and between 25.5% and 17.3% for the crown. Menéndez-Miguélez et al. (2021) analyzed the crown ratio (crown biomass/aboveground biomass, CR) and DBH<sup>2</sup>h (as a volume expression) relationship for different species. They found three different patterns: an increasing pattern, a constant one, and a decreasing pattern with tree size (DB-H<sup>2</sup>h). Pinus radiata followed the third trend, and this relationship was stabilized with a determinate value of DBH<sup>2</sup>h. These authors found a negative lineal model between these variables, which can be used in a wide range of *P. radiata* age plantation. Our results are only partially in accordance, although our plantations have not yet reached the constant value of CR probably because are young. The model which best explained this relationship was the exponential negative form:  $CR=0.3596 * e^{-0.01*v}$ , R<sup>2</sup>=0.98.

The relative amount of needles, branches and roots decreased with age, although it was more accentuated in the root. This decrease can be interpreted as a strategy that trees develop to survive in these soils with little effective depth. The development of biomass in the crown and root is crucial in young plantations, increasing the probability of survival at the development of stand (Cuong et al., 2020). Particularly in the crown, which is characterized by the size and length of the branches, it is partially associated with the available space for growth. Therefore, the smaller change in the proportion of needles and branches may be due to the lower density in the 21-year-old sites.

Root biomass represented a considerable proportion of the total tree biomass (17%-25%), averaging 21% during the whole age sequence. In C inventories, the estimation of the root biomass is crucial as it is a significant fraction of the total biomass and contributes to SOC since it remains on the site after harvesting. Given that sampling is complex and time consuming, it is usual to estimate root biomass from the RB/SB ratio by taking a factor of 0.20 (Brown et al., 1993). In this work, the RB/SB ratio decreased with the age of the stand, from 0.53 to 0.27, due to the strong accumulation of biomass in the stem. Laclau (2003b) found minor changes and the values were of 0.25 and 0.23, for 10- and 20-year-old P. ponderosa plantations. Our results are in partial agreement with Peichl & Arain (2007), who reported that root biomass decreased with age until the stand reached a constant RB/SB. We cannot indicate that the root/stem ratio is constant. This is possibly due to the fact that the age range studied was narrower than that indicated by the authors. Our result contrasts with Vanninen et al. (1996), who reported that the subterranean component was quite independent of tree age. The largest proportion of root biomass contributes to improve the absorption of water and nutrients in dry environments and to provide better support on sites with shallow soil (Peri et al., 2010). Considering that there are no previous reports of RB/SB ratio in the region, these values can be considered a first approximation, given that the sampling was conducted in a single site.

#### Soil organic carbon

The vertical SOC distribution described in our study is usually reported in the literature describing a decrease in C concentration along with profile depth (Zhang et al., 2019; Cuong et al., 2020) and it is attributed to the organic inputs that occur from the accumulation of the forest floor (Jobbágy & Jackson; 2003; Liu et al., 2016). We did not find a clear relationship between SOC concentration or SOC stock in each layer and age plantations. This is probably due to variability between sites and high SOC level in the short age range of plantations replacing grasslands. Rodríguez et al. (2015), for the same region, reported SOC concentration similar to our study and did not report significant differences in the SOC when comparing natural grasslands and a mixed forest plantation in the first cycle. Changes in the SOC content as an effect of the permanence of the plantation has been extensively analyzed in the scientific literature. Contrasting results have been reported on this aspect since they depend on the previous use and the type of soil. While some studies indicated that no significant increases in SOC are observed with stand age (Paul et al., 2002; Peichl & Arain, 2006), others reported significant increase in the first decade after planting (Cuong et al., 2020), whereas Pérez-Cruzado (2011) and Smal et al. (2019) reported gains starting at 10-15 years of age. Since the age sequence of our stands was not replicated and only represents the growth patterns of a stand over time, the relationship between SOC and stand age should be considered in accordance with these limitations (Peichl & Arain, 2006).

#### System carbon stock

C-stock in the plantation biomass was strongly associated with plantation age which is consistent with Peichl & Arain, (2006) and Zhang et al. (2019). The C-stock values in the tree biomass ranged from 26.7 to 105.2 Mg ha<sup>-1</sup> in the youngest and in the oldest plantation, respectively. This means an increase of 78.5 Mg ha<sup>-1</sup> (increased four times). These amounts are higher than those in Li et al. (2011) for 8 (2200 tree ha<sup>-1</sup>) and 19 (975 tree ha<sup>-1</sup>) year old *P. ko*raiensis plantations, where C-stock values were 0.75 Mg ha<sup>-1</sup> and 20.6 Mg ha<sup>-1</sup> respectively. However, Olmedo et al. (2020) reported a C-stock in the plantation biomass of 213 Mg C ha<sup>-1</sup> for a 20-24-year-old rotation, which doubles the value reported in this study at 21-year-old. As the authors indicated, changes in management (density) and rotation length could affect C storage. C-stock accumulation rate in aboveground biomass was 2.0, 4.3, 4.4, 4.5 and 4.5 Mg C ha<sup>-1</sup> year<sup>-1</sup>, for 9-, 13-, 15-, 19- and 21- year old plantations, respectively. These values are in a similar range to those reported by Balboa-Murias et al. (2006), which ranged at age 30 from 3.4 Mg C ha<sup>-1</sup> year<sup>-1</sup> to 5.9 Mg C ha<sup>-1</sup> year<sup>-1</sup>, in different sites and managements. Olmedo et al. (2020) reported values of 9.0 and 8.8 Mg C ha<sup>-1</sup> year<sup>-1</sup> for 20- and 24- year old plantations respectively, in contrast with our values that are almost the half. We believe that, in addition to forest management, the genetic material used, and site quality, may explain these differences.

Forest floor is a key component in C-stock dynamics, acting as a source of organic matter and a medium-term nutrient reservoir (Vesterdal et al., 2002), but it represents only approximately 3% of the total C stock at the system level. The C-stock amount stored in the forest floor and understory is lower than that reported from Merino et al. (2003) who found values of 26.7 and 37.8 Mg ha<sup>-1</sup> for 16-and 20-year-old plantations, respectively. On the contrary, these values were even higher than those reported by Li et al. (2011) for 8-year-old (3.14 Mg C ha<sup>-1</sup>) and 19-year-old (4.71 Mg C ha<sup>-1</sup>) plantations. The values found in this study were similar to those reported by Olmedo et al.

(2020). These variations can be explained by the very different climates, soils, and topographies. In addition to the environmental factors, the forest floor and understory C stock amount is also affected by forest management, age and canopy cover that can alter the availability of light, water, and nutrients (Zhao et al., 2014. In our study no relation was found between plantation age and C-stock in the forest floor and understory (r=0.02; p>0.05), in contrast to Zhang et al. (2019) and Smal et al. (2019). While in the 9-year-old stand most C-stock was derived from underground development owing to the lower density and lower tree cover, in the 13-, 19- and 21-year-old stands, C-stock was provided by the tree biomass.

SOC<sub>meq</sub> stock in the 0-50 cm layer was not associated with stand age and was similar (p>0.05) in the 9- and 21year-old stands. This is contrary to what was reported by Smal et al. (2019), Lei et al. (2019) and Cuong et al. (2020) for plantations of Pinus elliottii, P. sylvestris and Acacia mangium. The difference may be attributed to variations in the environment (climate and soil texture), species, and management applied to previous uses and even to the older age range of the stands in which the studies were conducted (Jandl et al., 2007). Although the age range in this study was lower, our results agree with Peichl & Arain (2006), who indicated that SOC was independent of stand age in a chronosequence of Pinus strobus L. 2- to 65-year-old. We observed a decrease in SOC<sub>meq</sub> stocks from 9 to 13 years of age. Li et al. (2011) in a Pinus chronosequence study of 2- to 35-year-old, attributed the initial losses to the change of land use from grassland to plantation, also mentioned in Pérez-Cruzado (2011). We cannot explain the reduction of SOC since the plantations are in a stage post-establishment. Laclau (2003a) stated that although soil carbon initially decreased when the natural grassland was replaced, the growth of the plantation generated a biological activity in deeper horizons that, with time, compensated for the initial losses. However, the greater the initial loss, the longer it will take to restore soil carbon. Conifers produce litter that contains more lignin, which decomposes more slowly (Chae et al., 2019). For this reason, the SOC's input is lower than grass's, and afforestation of grasslands with evergreen coniferous trees leads to an initial loss of SOC (Hou et al., 2020). This behavior was not clearly observed in our study since no significant changes were observed in the SOC<sub>meq</sub> stock in the 9- and 21-year-old stands, may be due to the existing variability between the soils was so great that it could not be reflected.

In the Pampas region of Argentina, other studies have analyzed changes in the concentration and SOC amounts across different land uses that replaced grasslands. Zalba & Peinemann (1987) reported higher SOC levels in forest plantations (coniferous and broadleaved) aged 30-50 years, compared to herbaceous vegetation. Berhongaray et al. (2013) reported average SOC stock of up to 1 meter of 131 Mg ha<sup>-1</sup> in forested sites. Forestations (>30 years old) significantly increased SOC content up to 1 meter compared to herbaceous vegetation and these effects were found in all soil types that included a large working area. Our results show that C-stock contents in the 0-50 cm (average of all age: 196.4 Mg ha<sup>-1</sup>) were higher than that reported by the authors. When we compare the average  $SOC_{meq}$  stock at 5 natural grassland sites adjacent to the sampled plantations (181.2  $\pm$ 11.2 Mg ha<sup>-1</sup>, unpublished data), we can see that there is a difference of 15 Mg C ha<sup>-1</sup> in favor of the plantations. The narrow age range may have conditioned the detection of changes in the SOC accumulation. There are few commercial plantations of P. radiata aged > 20-22 years in SE Buenos Aires; however, further studies should include older plantations to verify this trend. Another aspect to be considered is that only one site per age was available in this study, thus limiting a definite assessment of the age effect. These data would support what Alvarez et al. (2021) suggested, that it is possible that soils have not yet reached C-stock saturation on forestation sites that replaced grasslands, and thus have the potential to increase the C-stock sequestering capacity of these systems.

The C stock at the system level in the 21-year-old stand was 19% higher (51 Mg C ha<sup>-1</sup>) than in the 9-year-old stand and did not represent a significant increase. An average of 69% of the C-stock found was in the soil, 28% in tree biomass (97% in soil and tree biomass) and 3% in the forest floor and understory; similar tendency was reported by Samuelson et al. (2014) and Cuong et al. (2020). The increase in the proportion of C plantation in relation to the total C of the system (Fig. 5) until 19-year-old, was accompanied by a reduction in the proportion of C in the soil. The lower proportion in the 21-year-old plantation compared to the 19-year-old is explained by a higher carbon content in the soil in the 21-year-old stand.

Our study indicates that the *P. radiata* plantations biomass that replace natural grasslands in non-agricultural soils is the main contributor to C uptake, while the soil is an important C reservoir without variations in the age range studied. We think that there is a greater potential for C sequestration in agricultural land, in areas with less slope and deeper soils. These lands have gone through 100 years of agricultural use with an intensification process in the last 20 years, which have caused degradation and erosion processes and their consequent loss of SOC (Sfeir, 2015).

# Conclusions

This work provides equations for the calculation of different biomass components of *P. radiata* plantations in the sierras de Tandil chain in SE of Buenos Aires province, Argentina, thus serving as a tool for the C-stock estimation in this area. Beyond its practical use, this data also provides a basis for understanding the C accumulation patterns and distribution in the different forest plantation components, thus being a valuable contribution to regional and global studies. For the age range studied, the total tree, needles, branches, stem, and root biomass increased with the age of the stand; the proportion of stem biomass increased with age; C-stock in the plantation biomass was associated with stand age while it was not related to  $SOC_{meq}$  or C-stock in the forest floor and understory. Consequently, there was no relationship between stand age and the amount of C-stock in the system. At the system level, the soil component was the main C reservoir. Indeed, it would be convenient to incorporate sites in older stages to analyze variations to the observed trends, particularly in the forest floor and in the soil component. Analyses should also be implemented in other soil types, such as coastal zones, with sandy soil, probably resulting in different patterns of C accumulation in the components of the system.

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## **Authors' contributions**

Conceptualization: A. M. Lupi Data curation: P. Ferrere Formal analysis: P. Ferrere, A. M. Lupi Funding acquisition: P. Ferrere, A. M. Lupi Investigation: P. Ferrere, A. M. Lupi Methodology: P. Ferrere, A. M. Lupi Project administration: P. Ferrere, A. M. Lupi Resources: Not applicable Software: Not applicable Supervision: Not applicable Validation: Not applicable Visualization: P. Ferrere, A. M. Lupi Writing – original draft: P. Ferrere, A. M. Lupi Writing – review & editing: P. Ferrere, A. M. Lupi

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