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Allometric Equation to Estimate the Belowground Biomass of *Abies religiosa* Kunth Schltdl. et Cham

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Abstract: The use of allometric tools allows estimating the biomass of a plant species with greater precision. Tree roots are the least studied part of the vegetation due to the difficulty in obtaining complete root systems. For this reason, the objective was to generate an allometric model to estimate the amount of biomass accumulated in the root system of *Abies religiosa* trees in El Chico National Park, Hidalgo. For this purpose, 61 trees of *A. religiosa* were extracted from 0.06 to 3.56 m in height, using a backpack sprayer of pressurized water that detached the soil from the root. The response variable was root fresh weight and the independent variables were height, diameter and age+1, according to the fit of the models tested. Diameter was the variable with the best fit ($r^2 = 0.95 \pm 0.04$), obtaining the following equation PFR = (20.918 + D (2.4475)). The allometric model accurately estimates the below-ground biomass contained in the roots, which will allow us to obtain the complete biomass of an *A. religiosa* forest by having access to data on both the aerial and root components.

Keywords: allometry; root; carbon; El Chico National Park

1. Introduction

Over the last two decades, tropical land use change, especially deforestation and forest degradation, has accounted for 12% to 20% of global anthropogenic greenhouse gas (GHG) emissions [1]. Accurate assessment of forest carbon stocks is crucial for the successful implementation of climate change mitigation policies [2]. Currently quantifying forest carbon stocks accurately remains a global challenge [3].

The forest area of Mexico is approximately 65 million hectares, which represents one third of the national territory, 51% corresponds to temperate forests and 49% to tropical forests [4]. The forests of *Abies religiosa* Kunth Schltdl. et Cham. are important sinks of atmospheric carbon, it is an endemic conifer of Mexico [5] that is distributed mainly between 2400 and 3600 m altitude along the Transmexican Volcanic Belt. The main ecological function of the *A. religiosa* forest is as a preferred host for monarch butterfly populations that come to hibernate in Mexico [6], in addition to providing environmental services and serving as a reservoir of biodiversity of organisms such as saprotrophs and ectomycorrhizae [5]. Economically, it is important to highlight the use of wood as raw material for pulp and paper production and ecotourism for its impressive scenic beauty [7].

Studies on subterranean ecosystem processes are relatively scarce compared to those dealing with aboveground plant traits, since roots and rhizosphere are hidden in the soil [8]. Root studies are usually based on time-consuming and therefore costly methodologies [9]. Their limited information is due to the difficulty in obtaining complete root systems and because of the large amount of work involved in individual measurement, sometimes requiring heavy machines in their extraction [10–12], the same can be said of the reduced conceptual framework and existing terminology [13]. Consequently, it should be noted that the methodologies proposed so far show how complicated it is to avoid root losses and a certain percentage are broken during excavation [11]. Therefore, estimates of root biomass in trees are scarce, which leads to a lack of concise data on the global carbon stock found in a given environment [12].

In tree-dominated ecosystems, the aboveground biomass (BA) of vegetation is usually derived from soil survey data. Biometric measurements of trees are converted into biomass values using an empirical allometric model [14]. An allometric model is a tool that allows relating one or more easily measured variables (for example diameter at breast height, height, age) to estimate a variable that is difficult to measure (for example volume, biomass, carbon) [15].

The use of allometry tools allows estimating the biomass of a particular species with greater accuracy [13,15] are obtained from the extraction of trees and measurement of a data set for calibration [16,17]. The sample size for allometric equations for root biomass is small compared to those for aboveground biomass, due to the difficulties of excavating whole root systems [17], but which is necessary in all ecosystems to generate an allometric equation per species, allowing accurate biomass estimation by taking only a few data in a more accessible way and avoiding destructive sampling [16].

Several current publications are oriented to estimate the carbon stored in the different aerial components (trees, understory and necromass) in different types of vegetation [3,18,19], unfortunately the root component of carbon storage lacks estimates, Rojas-García and collaborators [20] conducted a database of allometric equations performed in Mexican forests, which resulted that of the 478 equations found in the literature only 5 belonged to tree roots, this limits the knowledge of the total amount of carbon stored in different ecosystems and the generation of climate change mitigation strategies. The objective of this study was to develop an allometric equation to estimate the belowground biomass of *A. religiosa.*

2. Materials and Methods

Study area and data collection

The work was carried out in El Chico National Park, located at the western end of the Sierra de Pachuca, located in the Transverse Neovolcanic Axis. It is situated between the extreme coordinates of 20°10'10'' to 20°13'25'' North latitude and 98°41'50'' to 98°46'02'' West longitude and covers an area of 2739 ha (Figure 1). Its altitudes range from 2600 to 3050 masl, it has a temperate-sub-humid climate with summer rains, an average annual <temperature between 12 and 18 °C, and the predominant soils are humic Cambisol, distric Regosol and humic Andosol of medium texture [21]. The 67% of the vegetation cover belongs to *Abies religiosa* forest [22].

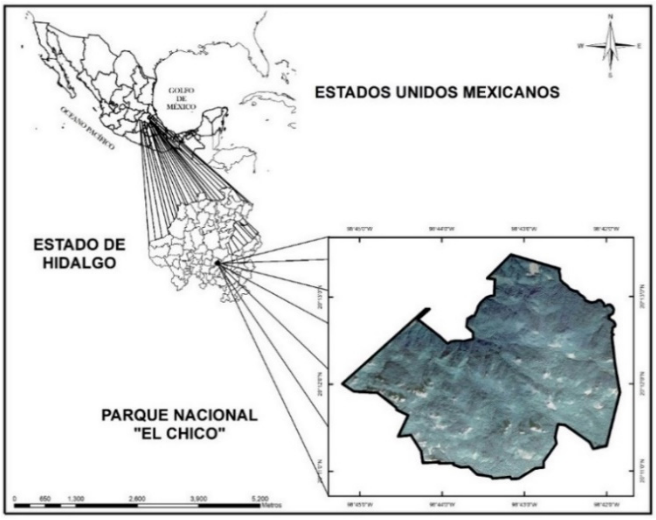

Figure 1. Geographic location of El Chico National Park Natural Protected Area.

Selection and extraction of A. religiosa trees

The sample size was calculated, considering a first sampling of 14 individuals assuming an error of 9% according to the initial sample, obtaining that 54 individuals should be sampled. To select the individuals to be extracted and to weigh the root biomass, we made rounds within the PNCH identifying spaces (clearings) in the forest wide enough to allow the natural regeneration of *A. religiosa*, then the trees were selected taking into account the following criteria: continuous heights from 6 to 356 cm, free of mechanical damage, alive and away from roads or gaps with compacted soil. The selected trees were prepared one day prior to extraction, which consisted of removing the moss and organic matter with a rake and adding sufficient water (an average of 80 liters) around the base of the individual to moisten and soften the soil in the area encompassing the root system, the next day with the use of a backpack sprayer of pressurized water, the soil was carefully removed from the root (Figure 2a), In order to ensure the complete extraction of the root system of each tree, each branch was located and followed its course with the rosator until the end of each root, which avoided the loss of the extremities as much as possible. In order to facilitate the work and avoid damaging the tree, a rope was tied in the middle part of the shaft to a wooden support in the form of a square fixed to the ground so that it remained suspended and in a vertical position to the ground so that it remained suspended and in a vertical position (Figure 2b) during the whole extraction process until the root system was completely free.

(a) (b)

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Figure 2. (**a**) Use of pressurized water backpack sprayer for root system extraction. (**b**) Wooden support to hold the tree to be extracted with the root system.

Variables evaluated

A total of 61 *A. religiosa* trees were extracted, seven more than the sample size obtained (54 samples) in order to increase the accuracy of the allometric model. Once the specimen was obtained, the root was carefully washed and left to air dry for approximately 2 hours [23]. The variables evaluated in the field were the following: diameter at the base, total height, age, fresh weight of the whole tree, fresh weight of the root, number of root branches and diameter of the main root. Diameters were measured with a Mitutoyo® digital vernier model CD-6 "CS with millimeter precision, height was measured with a 5-meter Truper® measuring tape graduated in mm, age was estimated by counting the number of whorls per individual since these are produced at a rate of one whorl per year in *A. religiosa* [24], the fresh weights of the whole tree and the root component were obtained with a BIOBASE® scale model BE16001. Subsequently, the aerial part and the root of each tree were placed separately in paper bags previously labeled and taken to the Seeds and Germplasm Laboratory of the Institute of Agricultural Sciences-UAEH for drying in the GRIEVE® model LW-201C oven at a temperature of 80 °C until constant weight was reached; the dry weight of the whole tree and of the root alone was recorded.

Data analysis

To determine the allometric model, multiple regression was used considering a potential distribution (a + (xb)n) of the data (n = variables), since it was the best fit ($r^2 = 0.93 \pm 0.04$). The response variable was root fresh weight (RFW), which was used to calculate biomass; the independent variables were total height, diameter at base and age+1, according to the fit to the potential model. The minimum predictive model was generated using Markov Chains (5000 steps by bootstrap) and the confidence interval was established for 50 iterations. The Akaike index (AIC) was the reliability indicator to determine the best model. The model was tested with 1000 random points to determine the resulting linear correlation between observed and expected fresh weight values [25].

The hypothesis of normality in the distribution of the residuals was verified with Pearson's Chi-square test, to test the hypothesis of homogeneity of variances, using the Lavene test [26]. Likewise, to ensure that the groups are morphologically distinct, a Cluster analysis was performed considering total height, diameter at base, age, number of root branches, fresh and dry weight of the tree and fresh and dry weight of the root, using Ward's method based on Euclidean distances. These groups were contrasted by Discriminant Function Analysis, after testing for multinormality using Mardia's test [27].

3. Results

The data obtained in the field from the 61 trees of *A. religiosa* extracted with the root system were subjected to multiple regression analysis to obtain the variables that best fit to generate the allometric model, the variables were: diameter at the base (DB), where the data obtained ranged from 0. 08 to 6.28 cm and total height (TH), which ranged from 0.06 to 3.56 m. The height variable was the main index when selecting the trees to be extracted, since starting with the first tree, we looked for specimens that were increasing in height, avoiding sampling individuals of the same height. Regarding the root biomass of *A. religiosa*, it varied according to the following factors: trees of small total height presented greater root biomass than trees of medium height, this is related to the fact that their ecological conditions of development differ, such as soil humidity, availability of space, competition for solar radiation, among others, the results obtained are in the range of 0.03 to 0.525 kg of root biomass per tree.

The equation generated in this study for the estimation of root biomass in *A. religiosa* trees can be used in other forests of the same species that meet certain conditions, such as the same type of soil, climate with similar ranges of temperature, precipitation and altitude above sea level, mainly. This

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model can be used in all stages of development of *A. religiosa* trees without increasing the range of error.

If the intention is to estimate the amount of root biomass present in an oyamel forest that complies with the ecological conditions, the allometric model can be applied by simply measuring the variables of total height and diameter at the base of the tree, which optimizes costs, time and labor in comparison with the total extraction of the individual. Rojas García et al. [20] mention that, to estimate the aerial biomass, the main variables considered to generate an allometric model are the total height and normal diameter of the tree, which results in that the same independent variables are mainly used to estimate the aerial and root biomass, which allows a better estimation of the possible variation due to ecological conditions.

The minimum model with the best fit and significantly less error was the one based on diameter, so this model was chosen. The equation proposed for *A. religiosa* is:

$$RFW = (20.918 + D\,(2.4475)).$$

where:

RFW=Root fresh weight (g)

D=Diameter (cm)

Model testing generated significant regression with fit 80.9% of cases ($F(1,998)=5058$, $p < 0.0001$, $r^2 = 0.8352$).

Confidence intervals indicate that models based on height+age, diameter+age, height+diameter and the full model did not differ from each other (Table 1), while models with single variables height, diameter and age differed.

Table 1. Confidence intervals of models applied to the variables of *A. religiosa* in the sapling stage.

	Akaike Index						
	HM	**MD**	**MA**	**MHD**	**MHA**	**MDA**	**MHDA**
Average	0.530	0.499	0.513	**0.595**	0.593	0.563	0.595
Standard deviation	0.007	0.007	0.006	**0.007**	0.007	0.007	0.007
Minimum	0.522	0.492	0.507	**0.588**	0.587	0.556	0.588
Maximum	0.537	0.506	0.520	**0.602**	0.600	0.570	0.602
Differences Sign.	b	c	b	**a**	a	a	a

HM=Height model; MD=Model diameter; MA=Model age; MHD=Model height diameter; MHA=Model height age; MDA=Model diameter age; MHDA=Model height diameter age.

Of all the variables evaluated, the number of root branches and diameter of the main root were not used in the development of the allometric model. According to the attributes evaluated, the model was able to retain an extrapolation rate of the biomass contained in the trees of *A. religiosa*.

The clustering analysis showed four groups related to all the variables and an isolated tree (Figure 3). With these groups, a discriminant classification analysis was performed to determine the most important variables in the model indicated by * in Table 2. In all cases the Mahalanobis distances obtained were significantly different, associated with a single discriminant factor explaining more than 99.003% of the variance between groups and with an eigenvalue greater than 16. Subsequent assignment analyses allow us to assign 100% of the individuals with their corresponding category, so that in future work it will be possible to distinguish the biomass according to the height and diameter of the individuals.

The groups formed correspond, for the most part, with the stages of crop development (sapland, coppice, latizar and forest) [28]. In addition, it is possible to subdivide the fustal considering trees larger than 35 meters as possible seedlings, and in this case, with greater biomass. The grouping characteristics standing height, number of root branches, tree fresh weight, root fresh weight, tree dry weight and root dry weight (Table 2) had not been previously considered in the suggested categories,

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so the estimation models were less accurate, so the current proposal was much more accurate and allows extrapolation to other areas with similar forest cover in terms of species composition.

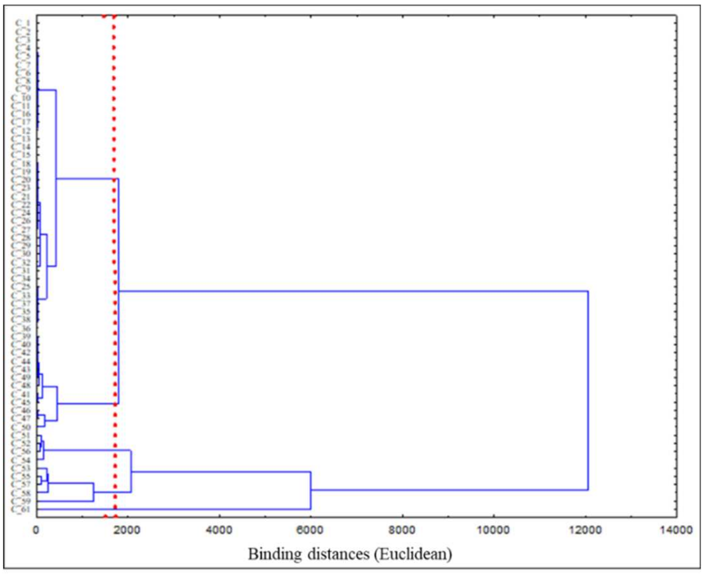

Figure 3. Amalgamation graph.

Table 2. Morphological characteristics of the groups formed.

Group	No.	Standing height (cm)*	Diameter at base (cm)	Age (years)	No. Root branching*	Fresh weight tree (g)*	Fresh weight root (g)*	Dry weight tree (g)*	Dry weight root (g)*
1	38	27,45 ± 17	0,42 ± 0,2	3,53 ± 2,2	6,11 ± 3,4	14,2 ± 16,6	3,54 ± 4,3	1,21 ± 1,8	0,36 ± 0,6
2	12	84,15 ± 13	1,19 ± 0,2	9,33 ± 0,8	10.25 ± 2,2	144,93 ± 61,2	37,7 ± 18,3	10,78 ± 36	2 ± 6,1
3	6	148,83 ± 17,9	1,95 ± 0,5	10,33 ± 1,4	15,33 ± 2,4	636,37 ± 218	168,62 ± 102,3	302,23 ± 89,2	78,12 ± 43,2
4	4	232,38 ± 47,8	3,06 ± 0,6	15 ± 1,6	20 ± 2	1366,53 ± 532,2	366,5 ± 140,2	753,3 ± 296,1	193,2 ± 70,4
5	1	356.0	6.285	19	16	3852.40	960.3	2065.34	525.3

By applying the equation obtained, the database was generated in .xlsx format, which requires entering the diameter at the base and total height of the tree of interest to obtain the root biomass, this allometric model can be applied in forests of *A. religiosa* with similar conditions to soil type, rainfall, height above sea level, average temperature, to name a few.

Quantifying the biomass present in all the components of an ecosystem makes it possible to estimate the availability of resources, the carbon storage capacity and, in turn, the loss of carbon and the factors that cause it over time, in order to develop strategies for its conservation [29]. Among these strategies is the payment for ecosystem services, specifically carbon credits, which consist of developed countries buying Reduced Emission Certificates from developing countries in order to reduce the over-accumulation of greenhouse gases. In most of the projects that are within this carbon market only receive payment for the vegetation present in the aerial component (trees and shrubs) because as mentioned in this study it is complex to estimate the biomass of the soil component, however, it is necessary to quantify it in its entirety in order to generate greater benefits to the owners of the forests and it is here when the generation of allometric models is highly applicable and important.

4. Conclusions

The allometric model generated to estimate root biomass in *A. religiosa* forest was PFR = (20.918 x D2.4475), which can be used in forests with ecological conditions similar to those of El Chico National Park, Hidalgo. In this model the best fitting variables were diameter at the base of the tree, therefore, this equation is a reliable tool to quantify belowground biomass in ecosystem services inventories, which will facilitate to know in a global way the root biomass content of a forest.

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