

RESEARCH ARTICLE

Assessment of Root-Shoot Ratio, Biomass, and Carbon Sequestration of Chestnut-leaved Oak Seedling (*Quercus castaneifolia* C. A. Mey)

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ABSTRACT

One of the most important ways to reduce atmospheric carbon is the carbon sequestration by trees. Chestnut-leaved oak (*Quercus castaneifolia* C. A. Mey) is one of the most important native oaks of Iran distributed in the Hyrcanian Forests. The pure and mixed stands of it cover about 6.5% of these forests. In this study, carbon sequestration of chestnut-leaved oak seedlings was evaluated by using some morphological characteristics of the root and shoot. For this purpose, one hundred seedlings were sampled by method of Systematic-Random from the sowing bed on March 2022 in the Pylambra nursery at Guilan province. Seedlings are divided to three grades small, medium and large according to Root Collar Diameter (RCD). The biomass and carbon sequestration of chestnut-leaved oak seedling were calculated according to the basic density of its root and shoot. The Pearson's correlation coefficient was used for correlation detection between variables. The one-way analysis variance test at the 95% confidence level was used to recognize difference among biomass and carbon sequestration of three group of the oak seedlings. The results of correlation analysis showed that the root collar diameter (RCD) had the strongest correlation with other morphological characteristics. The amount of the basic density for the root and shoot of the oak seedling was obtained about 0.57 g/cm³ which is the same for both of them. The amount of the biomass and carbon sequestration of the root was obtained more than shoot at the small and medium seedlings, whereas in large seedling was the same. In general, by increasing the size of seedling the biomass and carbon sequestration increased.

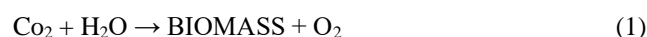
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1. Introduction

Carbon is the most important of greenhouse gas that in recent decades increasing of it has been caused earth warming in the atmosphere. Warming has damaging effects on lives and was caused destruction of natural ecosystems, occurrence of drought, climatic and ecological imbalance. Carbon sequestration in plant biomass and soils under the biomass is the most simple and cheapest possible way to reduce levels of this atmospheric gas.

As part of the carbon cycle, trees transform carbon dioxide to biomass through photosynthesis (Equation 1) (Liu & Li, 2012).



This function is beneficial to humans because it counteracts emissions of carbon dioxide (CO₂), a greenhouse gas. Anthropogenic carbon emissions have caused a 40% increase in atmospheric CO₂ concentrations in the last century, a change which is known to be causing global warming (IPCC, 2013). Whole-plant biomass of juvenile trees will greatly improve the

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accuracy of current estimates of forest carbon stocks for proposed new areas of indigenous afforestation/reforestation (Marden et al., 2018). Carbon sequestration involves the removal of CO₂ from the atmosphere, aiming to reduce the greenhouse effect. Wood basic density is a direct part of this process. Carbon dioxide (CO₂) gas, one of the compounds released into the atmosphere by man, is produced in all parts of the planet, mainly by burning petroleum-derived fuels and by producing cement (75% of total emissions). CO₂ is one of the greenhouse gases by the absorption of thermal infrared light. Carbon becomes available to living beings through plants by photosynthesis, and because carbon is stored, it is often called fixed carbon. *Quercus* is the largest genus in the family *Fagaceae* with about 300-600 species. This genus includes evergreen and deciduous shrubs and trees extending from cold latitudes to tropical Asia and Americas. Chestnut-leaved oak (*Quercus castaneifolia* C. A. Mey) is one of the most important species of Iran's native oaks, distributed in the Hyrcanian Forests (Panahi et al., 2011). The aim of this study is to: i) to develop a model explaining the shoot and root dry weight of the oak seedling according to variation of the root collar diameter ii) to understand the role of the oak seedling quality on carbon sequestration.

2. Materials and Methods

2.1. Study Area

This study was done on the oak seedlings that grow in the Pylambra nursery. The location of the Pylambra nursery is Pareh Sar city in Guilan province, Iran (49° 4' E, 37° 36' N) (Figure 1).

According to the climatic data received from the meteorological station, the climate of the area is very humid; and the mean annual temperature is 16.5 °C and average precipitation is 2139.7 mm.

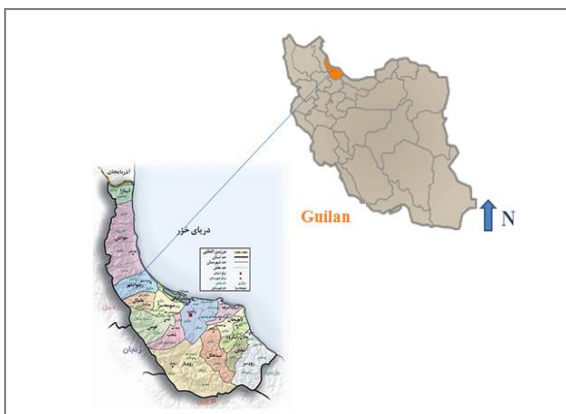


Figure 1. Locations of the study area in Guilan province, Iran.

2.2. Sampling Method of Seedlings

Samples were taken from chestnut-leaved oak seedlings grown within frames measuring 0.3 m x 0.3 m by Systematic

random sampling method (Figure 2). Sampling was done before the seedling transport to the planting fields on March 2022. More than 100 seedlings were sampled and then they transported to the forestry measuring lab in the faculty of natural resource at University of Guilan.

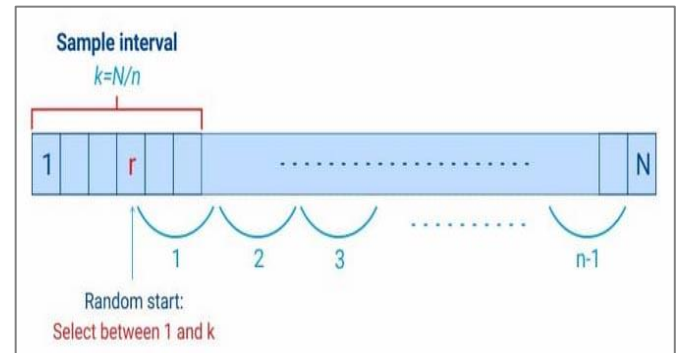


Figure 2. Systematic random sampling.

2.3. Measurement of Morphological Attributes

The oak seedlings were divided to three grades as small (3-6 cm), medium (6-9 cm) and large (9-12 cm) according to Root Collar Diameter (RCD). Then their morphological attributes were measured. Seedlings were divided into two sections: Shoot and Roots. Shoot height (SH), root collar diameter (RCD) were measured with accuracies 1 mm and 0.01 mm, respectively. Number of the First Order lateral Roots (FOLR) and Root length (RL) were measured and Root Volume (RV) was determined by water displacement method (Taherzadeh et al., 2014).

2.4. Measuring the Volume of Seedling Roots and Shoot

Seedling Roots have an irregularly shape. Therefore, using geometry is often difficult and complicated. The easiest way to measure root volume is using the water displacement method (Figure 3).

The volume of the oak seedlings was calculated by subtracting the volume of the water alone (V1) from the volume of the water plus the seedling roots (V2) (Equation 2).

$$V_{\text{root}} = V_2 - V_1 \quad (2)$$

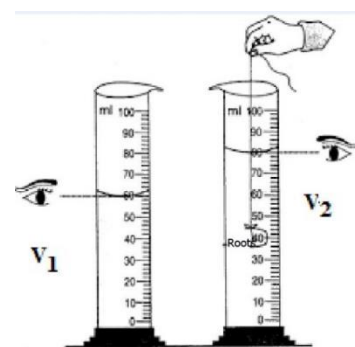


Figure 3. Measuring the volume of the seedling roots.

Unlike roots, the volume of each Seedling shoot was calculated according to the geometry Equation 3 (Kirby & Potvin, 2007).

$$V_{\text{shoot}} = \pi (RCD)^2 / 4 \times SH \times 0.5 \quad (3)$$

Where:

V_{shoot} = Seedling Shoot Volume (cm^3)

RCD = Root Collar Diameter (cm)

SH = Shoot height (cm)

2.5. Calculating Green Weight (GW) and Dry Weight (DW)

The green weight of a seedling is an estimate of the mass of the fresh seedling when it is alive.

Dry weight represents the mass of the wood in the seedling when dried in an oven, so the moisture is removed. For this purpose, at first the fresh weights of each part were measured. Then dry weight of them was measured after drying into Oven at 80 °C for 24 h (Taherzadeh et al., 2014).

2.6. Calculating Basic Density and Biomass of the Seedling

Basic density is the relationship between absolutely dry mass and saturated volume of seedling.

Basic density was determined by the maximum moisture content method reported and calculated as (Peichl & Arain, 2006):

$$BD = DW / V_s \quad (4)$$

BD = Basic density (g/cm^3)

DW = Dry Weight (g)

V_s = Saturated volume (cm^3)

$$B = V \times BD \quad (5)$$

B = Seedling Biomass (g)

V = Seedling Volume (cm^3)

2.7. Calculating Carbon Storage (C)

Carbon storage is the amount of carbon in the shoot and roots of the seedling. This is the total amount of carbon that is captured from the atmosphere during photosynthesis as well as the amount of carbon sequestered by the seedling. From experiments, scientists have found that about 50 per cent of biomass is carbon (Birdsey, 1992).

$$C = 0.50 \times B \quad (6)$$

C = Carbon storage of Seedling (g)

B = Biomass of Seedling (g)

$$CS = C \times 3.67 \quad (7)$$

CS = CO₂ Sequestration by a seedling (g)

2.8. Statistical Analysis

Grading is done on the basis of Root Collar Diameter (RCD). The accuracy of the grading tested by the Discriminant analysis. Kolmogorov-Smirnov test was used to verify the normality of data distribution and the Levene's test was used to evaluate the homogeneity of variances. Differences between mean of morphological attributes in diameters grades were tested using analysis of variance (one-way ANOVA). Wherever grades means were significant, the Duncan's post hoc test was carried out to compare the means. These analyses were conducted with the SPSS software (Version 19).

3. Results and Discussion

The age of all the oak seedlings was similar at the time of studying, but there were initial and significant differences in the above-ground metrics among them. The diameter class distribution of the oak seedling in the Pylambra nursery exhibited a tendency towards a bell curve distribution (Figure 4). The classes with most abundant individuals were 6-9 cm (medium) and 9-12 cm (large) with respectively 50 and 30% of individuals. Table 1 shows the descriptive statistics of the morphological characteristics of the oak seedlings.

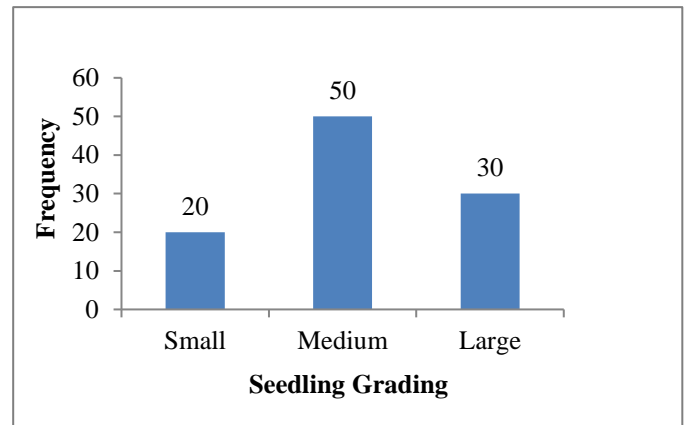


Figure 4. The diameter class distribution of the oak seedling in the Pylambra nursery.

Table 1. Descriptive statistics of the morphological characteristics of chestnut-leaved oak seedlings.

Seedling attributes	Mean±sd	Minimum	Maximum
Root collar diameter (mm)	7.3±0.4	3	14.8
Shoot height (mm)	556.09±20	66	1279
Shoot dry weight (g)	6.67±0.25	1	22.24
Root length (mm)	393±3	139	730
Number of the first order lateral roots	5.78±0.2	0	22
Root volume (cm ³)	10.25±0.5	2	65
Root fresh weight (g)	18.26±0.8	3.24	66.93
Root dry weight (g)	8.79±0.5	1.38	23.95

RCD is the base of the stem ends and the roots begin. Until recently, the standard minimum root collar diameter has been about 3.2 mm. Now, the industry standard is moving up. International Paper has increased its minimum standard to 3.8 mm or more in diameter. And the trend is in the direction of even larger seedlings sometimes up to 5.5 mm or more for special site conditions. The advantages of large-diameter loblolly pine seedlings are many, according to research on seedlings ranging in size from 3.8 mm to more than 7 mm (South, 1998). RCD was used instead of DBH in the analysis of seedling allometry because it was not expected to reach DBH height (1.3 m).

In this study, the RCD range for the oak seedling obtained between 3 mm and 12 mm. Results showed among the seedling characteristics, root collar diameter was most correlated with

others, therefore it was a good indicator of the seedling quality. Therefore, the grading is done on basic of on morphological attributes associated with this factor. Attributes of less than mean was considered as small, more than mean + Standard deviation as large and between these two states were considered medium. Finally, seedlings were graded into three grades (small, medium, large) based on the root collar diameter. Checking the grading accuracy on basic discriminant analysis showed that in both stage of sampling the probability more 90% grading is done correctly in each sampling stages.

The correlation coefficient and significance level between Root collar diameter (RCD) and Shoot height (SH), Shoot fresh weight (SFW), Shoot dry weight (SDW), Root volume (RV), Root fresh weight (RFW) and Root dry weight (RDW) was analysis using the Pearson's correlation coefficient (Table 2).

Table 2. Pearson's correlation coefficient and significance level of chestnut-leaved oak seedlings.

	RCD	SH	SFW	SDW	RV	RFW	RDW
RCD	1						
SH	0.595 **	1					
SFW	0.915 **	0.666 **	1				
SDW	0.88 **	0.549 *	0.954 **	1			
RV	0.837 **	0.593 *	0.895 **	0.800 **	1		
RFW	0.931 **	0.591 *	** 0.918	0.873 **	0.949 **	1	
RDW	0.917 **	0.460 *	0.898 **	0.904 **	0.890 **	0.978 **	1

** The significance level is 99%, * The significance level is 95%.

The most correlation obtained between RCD and weights of root and shoot of the oak seedling. A seedling with a large-diameter root collar can produce more wood in a shorter time than its average to smaller counterparts.

Comparison of carbon sequestration of shoot- and root biomass means were showed in three grades (Table 3).

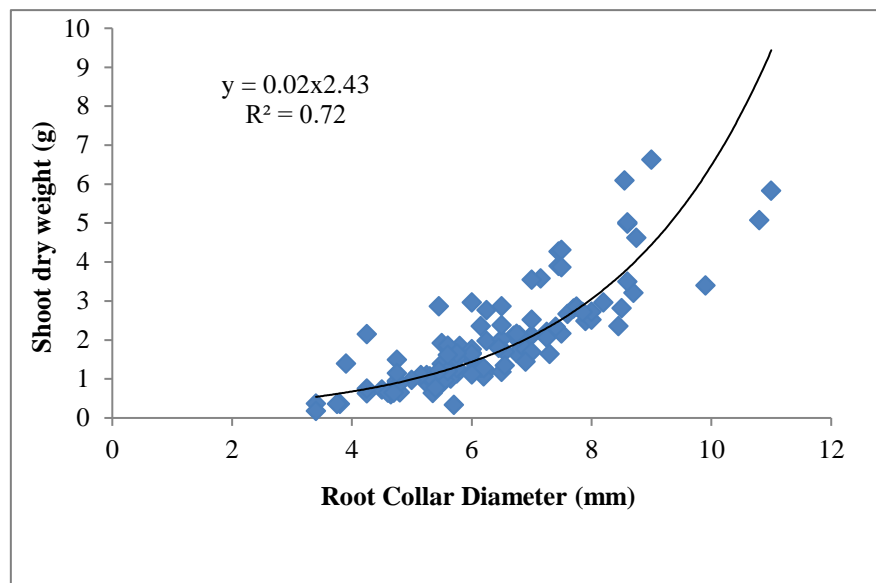
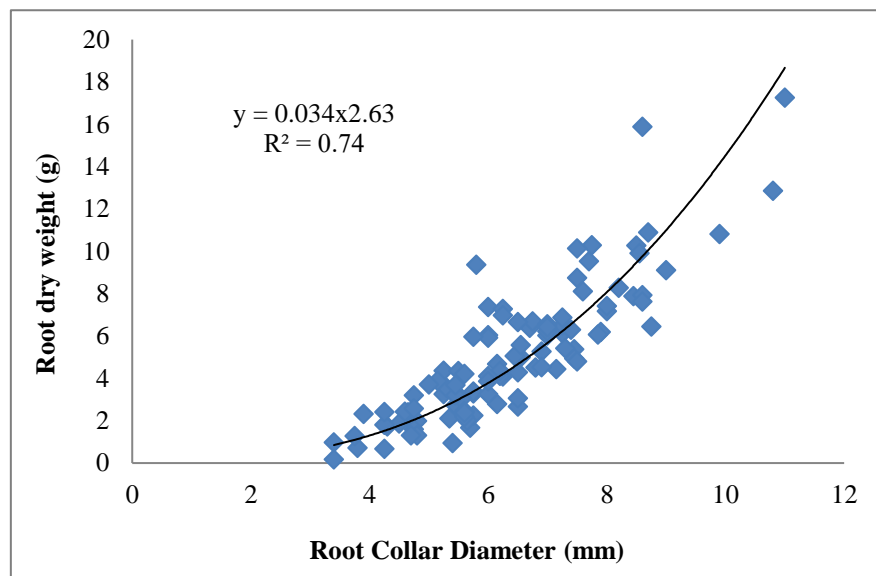
The highest carbon sequestration of shoot- and root biomass were in Large Seedlings with the numerical values of 8.66 and

8.28 g, respectively. The results indicate that larger grades seedlings are more capable in the production and use of carbohydrates and in nutrient uptake, and had grown more in the nursery, therefore probably have better performance in the field (Singh et al., 2011).

Figures 5 and 6 illustrate the trend of the shoot and root dry weights with an increase in RCD.

Table 3. Calculated parameters for three diameter classes of chestnut-leaved oak seedlings.

Parameters	Small Seedling (3-6 cm)	Medium Seedling (6-9 cm)	Large Seedling (9-12 cm)
Shoot volume (cm ³)	3.96	11.07	28.14
Shoot biomass (g)	2.26	6.31	16.04
Shoot carbon (g)	1.22	3.41	8.66
Root volume (cm ³)	7.21	14.66	26.92
Root biomass (g)	4.11	8.35	15.34
Root carbon (g)	2.22	4.51	8.28
Seedling volume (cm ³)	11.17	25.72	55.06
Seedling biomass (g)	6.37	14.67	31.38
Shoot/Root biomass ratio	0.55	0.75	1.04
Seedling carbon (g)	3.18	7.92	16.95
CO ₂ sequestration (g)	11.67	29.07	58.54

**Figure 5.** Power growth analysis of RCD and SDW.**Figure 6.** Power growth analysis of RCD and RDW.

The curves were plotted using the estimated shoot and root dry weight values in the power models. The curves have been obtained from estimated values of shoot and root dry weights.

Tree allometric equations are critical tools for determining tree volume, biomass and carbon stocks and have the potential to improve our understanding about carbon sequestration in woody vegetation to support the implementation of policies and mechanisms designed to mitigate climate change (Jara et al., 2014). The models for the prediction of biomass, though related to a specific case study are a tool of considerable utility for both ecological and silvicultural purposes (Marziliano et al., 2015).

Two-parameter power regression analysis was a good fit for the RCD and SDW data, with $R^2=0.72$ and also for the RCD and RDW data, with $R^2=0.74$.

4. Conclusion

Generally, in this study the RCD of chestnut-leaved oak seedling obtained more than the standard minimum root collar diameter at the same ages. The RCD detected a good indicator of the oak seedling quality and also has high correlation with other morphological attributes. The oak seedling was graded according to RCD into three grades (small, medium and large). The highest Carbon Sequestration of Shoot and Root biomass obtained in large seedlings. The power regression obtained a good fit model for the RCD and SDW data and also for RDW data.

Declaration

An earlier version of this article was presented at the 3rd International Congress on Engineering and Life Science at Karadeniz Technical University, Trabzon on 20-22 September, 2023.

Conflict of Interest

The authors declare that they have no conflict of interest.

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