

Article



# Assessment of Forest Biomass and Carbon Storage in Habitat 9340 *Quercus ilex* L. to Support Management Decisions for Climate Change Mitigation

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Abstract: The assessment of forest biomass has been a focus of research, aiming to understand matter-energy relationships in forest ecosystems and address forestry practice issues. In recent decades there has been increased societal interest in rational forest resource exploitation, necessitating accurate biomass estimation. In Greece there has been limited efforts in estimating forest biomass, and the current study focuses on habitat type 9340, "Quercus ilex and Quercus rotundifolia forests," located in the protected areas "GR1420004-Karla-Mavrovouni-Kefalovryso Velestinou-Neochori" and "GR1430001-Oros Pilio and Paraktia Thalassia Zoni (Mount Pilion and Coastal Sea Zone)" in the Natura 2000 network. The habitat falls within the thermo-Mediterranean zone and the study aims to estimate the biomass and carbon storage to contribute to sustainable EU forest strategies. Due to resource limitations, a generalized allometric equation was proposed as an alternative to traditional biomass estimation methods. The above-ground biomass per hectare was estimated, ranging from 16.10 to 205.27 Mg ha<sup>-1</sup> (mean 61.91 Mg ha<sup>-1</sup>). Furthermore, two approaches were used to estimate the total biomass in the habitat: regional averages based on spatial distribution and spatial interpolation using a geographic information system. The total estimated biomass for habitat 9340 is 183,505 Mg, with the carbon storage in standing dry biomass amounting to 83,725.25 Mg. Despite the absence of sampled biomass specimens, this study combines robust statistical techniques with published empirical values to provide a solid framework for estimating assimilated CO<sub>2</sub>. Sequestered CO<sub>2</sub> in the study area is estimated at 306,992.58 Mg. Therefore, the significant role of Quercus ilex L. in carbon storage in Mediterranean forest ecosystems is highlighted by sequestering a substantial amount of CO<sub>2</sub>.

**Keywords:** forest biomass; forest habitat; above-ground biomass; carbon storage; allometric equations; carbon sequestration

# 1. Introduction

The assessment of forest biomass has systematically engaged researchers for a period exceeding 70 years, with the goal of comprehending matter–energy relationships within forest ecosystems and addressing issues related to forestry practice (e.g., the spatial and temporal organization of forest stands). Concurrently, alongside researchers' interest in determining forest biomass, various societal groups in recent decades have focused their



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**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). attention on the more rational exploitation of forest resources, a prerequisite for which is the estimation of biomass in forest stands. Furthermore, Hall [1] delineates the diverse social and environmental benefits arising from the utilization of biomass as a means of energy production for the 21st century.

According to Johansson et al. [2], Hall [1], and the Intergovernmental Panel on Climate Change (IPCC) [3,4], three of the main reasons for the reassessment of the significance of forest biomass in research, political, and economic domains are:

- The direct relationship of organic matter to the carbon cycle in forest ecosystems and, by extension, to the mechanisms of the Kyoto Protocol (KP);
- The potential use of forest residues from forestry operations (branches, foliage, bark, etc.) as a source of bioenergy;
- The determination of the productivity of terrestrial ecosystems, which is directly linked to the flow of energy, nutrient cycles, and the state of the natural environment.

Today, an in-depth study of the impact of increasing atmospheric greenhouse gas concentrations on forest ecosystems, through the process of photosynthesis, necessitates the estimation of their above-ground and below-ground biomass [5,6]. Biomass estimation is one of the key variables in comprehending the photosynthesis process. A result of the conducted research projects on forest biomass estimation is the development of relevant databases, and publications at regional, national, and international levels. The utilization of an appropriate and stringent methodology to precisely ascertain forest biomass is of the utmost significance in the realm of ecological research and environmental sustainability, and several approaches have been introduced over time [7].

Kittredge [8] is considered a pioneer in biomass estimation for oak stands in Great Britain, while Ovington [9] and Ovington and Madgwick [10] estimated biomass in Scots pine stands in the same country. Studies on forest biomass estimation were conducted in Japan during the 1950s (for a comprehensive bibliography, refer to Satoo and Madgwick) [11]. Towards the end of the 1960s, global efforts were made to quantify the biomass of forest ecosystems through the International Biological Program, funded by UNESCO [12]. Subsequently, the oil crisis of 1973 led to the reconsideration of the use of renewable resources, such as forest wood, as sources of energy. This resulted in various research programs aimed at quantifying and potentially utilizing the biomass of forest stands.

In summary, the following works are briefly mentioned. Ter-Mikaelian and Korzukhin [13] compiled a considerable number of equations used for estimating tree biomass in North America. Jenkins et al. [14] further enhanced the aforementioned work by introducing equations published after 1997, as well as those developed for forest species found in Canada. In Australia, the National Greenhouse Gas Inventory commissioned Eamus et al. [15] and Keith et al. [16] to conduct a literature review, in order to develop a biomass equation database for the forest species present in the country. At the European level, Zianis et al. [17] compiled 607 biomass equations from works published in scientific journals, conference proceedings, scientific yearbooks, etc., within the framework of the COST E21 program, funded by the European Union [18]. Subsequently, Muukkonen and Mäkipää [19] added 188 new equations, thus covering more countries in the European continent. Recently, Falster et al. [20] published a database for biomass and allometric biomass equations for various woody plants titled "BAAD."

In Greece, few efforts have been made by researchers to estimate forest biomass. The impact of forest fires on carbon balance becomes indirectly apparent [21], emphasizing the necessity to preserve the ecosystems in the broadleaved evergreen zone in order to lower the concentration of several pollutants, especially in overpopulated areas [22]. In a Scots pine forest in Kassandra (Chalkidiki, northern Greece) the above-ground biomass (AGB) in plots of Aleppo pine (*P. halepensis* Mill.) was estimated from 9.51 Mg ha<sup>-1</sup> (for 23-year-old trees) to 94.12 Mg ha<sup>-1</sup> (for trees older than 100 years) [23]. Further west of Chalkidiki, Zianis and Mencuccini [24] developed allometric biomass equations for beech trees (*Fagus moesiaca* Cz.) in the municipal forest of Naoussa (Mount Vermio), while Zerva et al. [25] estimated the biomass of the root system in oak stands on Mount Paiko. Furthermore, the

crown fuel biomass of Aleppo pine was estimated via allometric equations, based on the sampling of 40 individuals in a forest in Evia [26], while in Taxiarhis, Chalkidiki, Zianis et al. [27] developed allometric biomass equations for *Quercus frainetto* Ten. trees.

In the present study, habitat type 9340 "Quercus ilex and Quercus rotundifolia forests," a habitat type in Annex I of Directive 92/43/EEC on the Natura 2000 network, was studied. Habitat type 9340 includes forests dominated by Quercus ilex or Q. rotundifolia (Quercus rotundifolia refers to western Europe). Moreover, habitat 9340 is found in the thermo-Mediterranean zone. The form of the habitat can be forests or high shrubs, with evergreen sclerophylls. They are often degraded to arborescent matorral [28]. The dominant species in habitat 9340, Quercus ilex L., commonly known as holm oak, is a broadleaved evergreen tree or shrub, which can reach 25 m, native to the central-western Mediterranean basin. The holm oak tree can thrive in many different Mediterranean climates and in a wide range of soil types. Q. ilex forests have great value as biodiversity reserves and are usually managed as coppice forests, producing mainly charcoal and firewood [29].

*Q. ilex* is vulnerable to drought stress. Negative effects, as co-factors of drought, could include air pollution and soil contamination [30]. Therefore, in the context of climate change and its predicted effects in the Mediterranean area, a reduction in holm oak forests is fore-seen [31,32]. The most important threats to and pressures on habitat 9340 include fire and fire suppression, landslides, anthropogenic reduction of habitat connectivity, cultivation, grazing, forest and plantation management and use, settlements and infrastructure, and erosion etc. [33].

In Greece, most of the once-dense holm oak forests were either converted into agricultural land, settlements and infrastructure, or were degraded and turned into coppice forests, which are easier to manage and exploit. Repeated fires and overgrazing by farm animals contributed to the degradation [34]. *Q. ilex* is a common element of the maquis in all floristic regions of Greece, particularly in locations in the mainland and the Ionian islands, with comparatively high precipitation, and in ravines. In the composition of the undergrowth, the species *Quercus coccifera*, *Erica arborea*, *Pistacia terebinthus*, *Arbutus andrachne*, *Arbutus unedo*, and *Phillyrea latifolia*, etc., also participate, to a lesser extent.

Focusing on the literature sources regarding habitat type 9340, it was found that there are no published studies on biomass estimation in this habitat. Nevertheless, several studies developing allometric equations were investigated regarding the dominant species in this habitat type, which is the holm oak (*Quercus ilex* L.). As reported by Canadell et al. [35], biomass equations for the aforementioned species have been developed in Italy by Susmel et al. [36], as well as by Leonardi and Rapp [37], and can be applied to trees with a diameter of 5–30 cm. Canadell et al. [35] developed equations for the same species in the Spanish region of Montseny, for trees with a breast height diameter (DBH) of up to 30 cm, or a measured diameter, for 50 cm above the ground, of up to 35 cm [35]. Rapp et al. [38] estimated the biomass of a holm oak stand (40 years old) in the Montpellier area, with a density of 3850 individuals per hectare, an average height of 4.7 m, and a mean DBH of 7.0 cm. Sabaté et al. [39] estimated the biomass expansion factor (BEF), the AGB per unit area, and the carbon content in the organic matter for various forest species in Catalonia. For various species in Spain, Ruiz-Peinado et al. [40] presented equations that relate the linear dimensions of trees (diameter and/or height) to the biomass of the trunk, branches, leaves, and roots, while Boulmane et al. [41] estimated the AGB of an evergreen oak forest in two experimental plots in the Moroccan Middle Atlas area.

The purpose of this paper is to estimate the biomass and carbon storage in habitat 9340, in order to assist decision making towards sustainable EU forest strategies [42]. Therefore, field measurements were carried out to estimate the biomass and carbon storage. Sampling plots were established in the study area, in which the dendrometric characteristics were recorded (diameter at breast height, tree average height). The Materials and Methods section provides a brief literature review of the research studies on biomass estimation in forest ecosystems and shrublands, both on a global scale and at a national level. Special emphasis is given to studies focusing on Mediterranean ecosystems, since *Quercus ilex* 

is of the utmost importance in the basin [43,44], and to the process of selecting the most appropriate method for AGB estimation. In this context, the estimation of biomass and carbon storage was carried out based on a generalized allometric equation. Finally, the results of the biomass and carbon storage estimation are presented for habitat 9340.

## 2. Materials and Methods

# 2.1. Study Area

The wider study area is located in Thessaly, Greece, and more specifically in the Regional Units of Magnesia and Larisa (Figure 1). It is characterized by a rich natural environment, as in the area the protected areas of "GR1420004–Karla–Mavrovouni–Kefalovryso Velestinou–Neochori" and "GR1430001–Oros Pilio and Paraktia Thalassia Zoni (Mount Pilion and Coastal Sea Zone)" in the Natura 2000 network are found. The areas, GR1420004 and GR1430001, have both been designated as a Special Area of Conservation (SAC). These areas are under the responsibility of the Management Unit of the Protected Areas of Thessaly (under the Natural Environment and Climate Change Agency (N.E.C.C.A.)).



Figure 1. Map depicting the Regional Units of Magnesia and Larisa.

GR1420004 and GR1430001, according to the Standard Data Form (SDF) for the areas in the Natura 2000 network, are characterized by a wide variety of habitat types in Annex I of Directive 92/43/EEC. One of the habitat types located in these areas is 9340 "*Quercus ilex* and *Quercus rotundifolia* forests". In GR1420004, habitat type 9340 covers an area of 1183.19 Ha and, in GR1430001, it covers an area of 2062.58 Ha (Figure 2).

The climate in the region is characterized as the Mediterranean type. It is a temperate climate type, with hot, dry summers and cool, wet winters. Habitat 9340 "*Quercus ilex forests*" probably originates from the remnants of larger areas, which have been reduced due to human activities (Figure 3). The main pressures–threats to the study area come from human activities, such as livestock farming, tourism, and development projects.



**Figure 2.** Map depicting the study area of habitat 9340 (official boundaries of habitat 9340 in yellow), located in the broader region of Magnesia and Larisa.



Figure 3. Quercus ilex in habitat 9340: (A) leaves and acorns, (B) typical habitat area, (C) sparse stands, (D) tall holm oak tree.

#### 2.2. Methodological Framework for the Field Measurements

In the context of recording the status of habitat 9340 and estimating the stand biomass, field measurements were carried out by establishing sampling plots (SPs) in the 9340 habitats in the GR1420004 and GR1430001 areas. The SPs were established in areas where habitat 9340 was found.

During the field survey, field measurements were carried out, in a total of 43 SPs. More specifically, 36 SPs were in GR1420004 and 7 SPs were in GR1430001. Each SP had a rectangle shape and an area of  $500 \text{ m}^2$ . The following data were obtained from each plot:

- The diameter at breast height of each tree (>6 cm), using a caliper;
- The average height of the trees in each diameter class (in meters);
- The altitude, slope, and aspect of each SP.

#### 2.3. Methods for Estimating Biomass in High Forests and Shrublands

The estimation of forest biomass is a time-consuming process that requires intensive fieldwork to collect the appropriate data and specialized personnel to generate useful information. In summary, several methods for estimating organic matter in forests are briefly mentioned below:

- The allometric regression equation, based on the development of empirical models, constitutes the primary approach for estimating biomass at the stand level. The application of empirical allometric equations, which link the dry organic matter of the tree (dependent variable) with one of its linear dimensions (stem diameter, height, etc.), or a combination thereof (independent variable or variables), is the most widespread approach for biomass estimation at the cluster level. These models are applied at tree scale in each cluster, and their sum yields the total biomass [45–47].
- The mean tree method, according to which individuals whose diameter approximates the quadratic mean diameter are selected for destructive biomass sampling. Subsequently, the average dry biomass of these trees is computed and multiplied by the total number of trees in the stand [48].
- The mean plot method, in which the biomass is estimated for several SPs, and in turn the average value (on an area basis) is calculated. Subsequently, this average is multiplied by the total area of forest stands [11,49].
- The biomass expansion factor (BEF) method is based on volume estimates of stands and their conversion to dry weight per plot area using a coefficient [50].
- The method involving generalized allometric equations. Generalized equations have been developed by Pastor et al. [51] for six species in the USA, for European beech trees by Zianis and Mencuccini [24], and for various European species by Muukkonen [52].

It should be noted that in recent years, the use of remote sensing data has been employed for estimating organic matter in forests. However, these data inherently rely on the application of the abovementioned methods to evaluate the results. An additional drawback to the remote sensing approach is that the recorded radiation is not directly related to biomass, but to a variable (e.g., tree height, crown density, crown diameter, etc.) that should be connected to organic matter based on empirical equations. The main advantage of remote sensing in estimating forest biomass derives from the fact that vast areas can be covered (depending on the spatial scale of the satellite data analysis) in a relatively short period and at a relatively reduced cost [53–55].

Regarding biomass estimation methods in shrublands, Ogawa and Kira [45] describe two methods (forest areas where the mean height of the dominant trees does not exceed 5 m). The first method is similar to the one applied in high forests (regression analysis). The dendrometric characteristics (base diameter, height, crown length, etc.) of a sample of trees are linked, through empirical statistical models, to the dry biomass. Then, the dendrometric characteristics are recorded for all the individuals within the study area, in order to apply the empirical models. The second method also involves the use of empirical models, but they are used on an area basis. The biomass (Mg ha<sup>-1</sup>) of a specific SP is calculated after the logging of all the individuals within the plots. Subsequently, the biomass of each SP is correlated, using empirical models, with "stand parameters" (such as trees per ha, circular plot area per ha, mean height, ground-cover percentage, etc.), which are considered independent variables. Finally, these models are applied to unlogged SPs, where the appropriate stand variables have been recorded to estimate the biomass at the forest level.

#### 2.4. Estimation of Biomass in Evergreen Broadleaved Forests

The harvestable biomass (HE) for three land cover classes is given by the following equation:

$$HE = b_0 + b_1 H + b_2 X_1 + b_3 X_2 \tag{1}$$

where  $1 \le H \le 3.5$  m represents the mean height of the dominant trees, and  $X_1$  and  $X_2$  are discontinuous variables that take certain values depending on the degree of ground cover [56]. Specifically, when the ground cover is less than 40% (sparse shrubs), then  $X_1 = 0$  and  $X_2 = 0$ . For loose forms and a ground cover degree of 41–70%,  $X_1 = 1$  and  $X_2 = 0$ , while for dense shrubs with a ground cover degree exceeding 70%,  $X_1 = 0$  and  $X_2 = 1$ . The coefficients in the equation were formulated as follows:  $b_0 = 5.7467$ ,  $b_1 = 2.4991$ ,  $b_2 = 4.7487$ ,  $b_3 = 8.1747$ . The site quality and the composition of forest species did not affect the coefficients in the equation.

#### 2.5. Selection of Biomass Estimation Method

The optimal method for biomass estimation in the study area would require the logging of several individuals, their drying, the recording of their biometric characteristics, and the development of appropriate statistical models (allometric equations). However, due to limited resources, the application of this method was not possible. As an alternative approach, the development of a generalized allometric equation (Section 2.2) was proposed, based on published empirical models from studies conducted in ecosystems dominated by *Quercus ilex* L. The published allometric equations are in the form:

$$M = aD^b \tag{2}$$

where M represents AGB, D represents the diameter at breast height (DBH) at 1.3 m above the ground, and a and b represent the pair of empirical coefficient values. In Table 1, the empirical values of coefficients a and b in the equation are presented, along with the bibliographic source from which they originated and the country for which they were developed. The minimum applicable diameter is 5 cm, while in all equations the upper diameter exceeds 30 cm. The coefficient of determination in all equations was above 90%.

**Table 1.** Published allometric equations in the form  $M = aD^b$ .

Reference	Area	a	b	Equation	Reference
Susmel et al. [36]	Italy	0.236	2.2791	$M = 0.236D^{2.2791}$	(3)
Ferres et al. [57]	Spain	0.2313	2.2662	$M = 0.2313D^{2.2662}$	(4)
Canadell et al. [35]	Spain	0.2269	2.207	$M = 0.2269 D^{2.207}$	(5)
Leonardi and Rapp [37]	Italy	0.2179	2.0513	$M = 0.2179 D^{2.0513}$	(6)
Rapp et al. [38]	France	0.4579	1.654	$M = 0.4579 D^{1.654}$	(7)
Sabaté (personal contact)	Catalonia	0.1646	2.2671	$M = 0.1646D^{2.2671}$	(8)
Ruiz-Peinado et al. [40]	Spain	0.3152	2	$M = 0.3152D^2$	(9)

In Figure 4, the predicted biomass values are shown for each of the equations in Table 1, for trees with a DBH within the range of 5–30 cm. The percentage difference in the biomass values for small-sized individuals (D = 5 cm) can reach up to 50% (6 kg predicted by Equation (6) and 9 kg by Equation (3)). For larger-sized individuals (D = 30 cm), the predicted values range from 127 to 535 kg.



**Figure 4.** Predicted biomass values from the published equations in Table 1,  $5 \le D \le 30$  cm.

The significant variation in the predicted AGB values in Figure 4 raises the question of selecting the less biased published allometric equations for the study area. An indirect way to assess the accuracy of the published equations is to compare the predicted biomass values per unit area against the standing timber volume of the 43 SPs that were established in the study area. The standing wood volume per hectare was estimated using the quadratic mean stem volume method. Thus, each one of the seven equations was applied to the diameter distribution of the 43 SPs, and the total above-ground dry biomass for each plot was calculated. Subsequently, the total biomass for each sampling plot was converted to a hectare basis (Table 2). The linear regressions of AGB (derived from the seven different equations) against the standing wood volume per hectare are presented in Figure 5.

**Table 2.** Above-ground dry biomass in Mg ha<sup>-1</sup> as calculated for the 43 SPs, based on the seven published equations from Table 1. The index number in the header of each column corresponds to the equation from Table 1.

Plot	Volume m <sup>3</sup> ha <sup>-1</sup>	$M_3 \ { m Mg} \ { m ha}^{-1}$	$M_4$ Mg ha $^{-1}$	$M_5$ Mg ha $^{-1}$	$M_6$ Mg ha $^{-1}$	$M_7$ Mg ha $^{-1}$	$M_8$ Mg ha $^{-1}$	M9 Mg ha−1
1	20.19	31.71	57.63	54.84	47.02	27.28	40.85	39.11
2	22.33	35.31	67.38	63.94	54.12	28.06	45.01	45.61
3	17.42	27.24	49.69	47.28	40.49	23.33	35.08	33.72
4	16.67	24.82	44.84	42.69	36.65	21.58	32.02	30.44
5	13.44	28.73	54.14	51.42	43.66	23.22	36.71	36.67
6	21.06	33.17	62.92	59.73	50.62	26.67	42.34	42.6
7	21.6	28.32	53.35	50.67	43.04	22.84	36.18	36.14
8	13.16	17.23	32.58	30.93	26.24	13.81	21.99	22.06
9	24.71	32.44	61.91	58.76	49.73	25.64	41.33	41.91
10	15.24	16.86	33.54	31.75	26.58	12.47	21.28	22.65
11	20.15	24.75	47.83	45.36	38.27	19.17	31.44	32.36
12	8.99	12.67	23.63	22.45	19.12	10.4	16.22	16.02
13	20.29	26.67	50.48	47.93	40.66	21.36	34.04	34.19
14	18.69	21.77	42.09	39.91	33.66	16.98	27.67	28.47
15	21.7	28.51	53.29	50.63	43.07	23.57	36.51	36.11

Table 2. Cont.

Plot	Volume	$M_3$	$M_4$	$M_5$	$M_6$	$M_7$	$M_8$	$M_9$
	m <sup>o</sup> ha <sup>-1</sup>	Mg ha <sup>-1</sup>						
16	20.87	27.18	50.13	47.67	40.71	22.82	34.9	34
17	19.78	30.88	59.16	56.13	47.45	24.32	39.31	40.04
18	13.75	17.54	30.97	29.52	25.5	15.85	22.74	21.05
19	23.12	28.94	54.74	51.97	44.07	23.48	36.97	37.07
20	18.39	23.27	43.79	41.58	35.31	19.03	29.75	29.66
21	23.48	30.62	56.63	53.84	45.94	25.56	39.29	38.4
22	20.19	31.71	57.63	54.84	47.02	27.28	40.85	39.11
23	22.33	35.32	67.38	63.94	54.12	28.06	45.01	45.61
24	17.42	27.25	49.69	47.28	40.49	23.33	35.08	33.72
25	38.99	78.63	74.32	61.75	38.41	27.10	53.03	48.19
26	132.51	289.65	273.32	225.37	137.31	91.47	195.04	171.07
27	68.61	149.65	141.67	118.56	75.06	54.92	101.07	94.68
28	47.12	108.11	102.37	85.75	54.43	40.07	73.03	68.71
29	59.79	130.64	123.61	103.20	65.14	48.27	88.19	82.18
30	12.89	32.09	30.56	26.27	17.87	15.87	21.79	23.10
31	113.15	200.51	189.56	157.64	98.27	69.63	135.25	123.38
32	60.66	104.00	98.67	83.43	54.29	42.69	70.39	69.12
33	137.84	312.52	295.99	248.24	158.17	118.17	211.16	199.99
34	80.36	210.70	199.32	166.24	104.34	74.77	142.21	131.25
35	160.77	284.43	268.27	220.74	133.72	87.67	191.45	166.27
36	155.79	283.70	267.65	220.45	133.90	88.32	190.99	166.63
37	17.13	44.04	41.91	35.93	24.23	20.87	29.89	31.22
38	20.27	53.75	51.07	43.49	28.86	24.01	36.43	36.99
39	24.73	71.73	68.07	57.57	37.47	29.46	48.55	47.71
40	22.67	58.95	56.08	47.97	32.16	27.11	40.00	41.33
41	34.53	85.19	80.91	68.70	45.17	36.33	57.71	57.69
42	185.70	235.10	221.96	183.43	112.38	75.75	158.38	140.25
43	46.79	114.88	108.79	91.17	57.96	43.04	77.61	73.23



Figure 5. Cont.



Volume (m<sup>3</sup> ha<sup>-1</sup>)

**Figure 5.** Regression of AGB in relation to STV. The estimation of the biomass in each diagram was derived from the respective equations in Table 1.

In all cases, the coefficient of determination indicates a strong relationship between the two variables (exceeding 87%), and all the biomass equations produced reasonable results. In other words, the predictions made by the empirical equations linearly increase in relation to the standing volume. Nevertheless, a closer examination of the slopes in Figure 5 reveals a difference in the absolute values of the above-ground biomass in relation to the standing tree volume. The criterion to select the most appropriate equation (or equations) for developing a generalized allometric equation was the mean value of the BEF. The BEF is defined as the fraction of above-ground biomass (ha<sup>-1</sup>) to the standing tree volume (STV) (ha<sup>-1</sup>) as follows [58,59]:

$$BEF = \frac{AGB[Mg ha^{-1}]}{STV[m^3 ha^{-1}]}$$
(10)

Sabaté et al. [39] estimated the mean value of the BEF for Catalan *Q. ilex* L. ecosystems to be 1.28 Mg m<sup>-3</sup> (sd = 0.15). Based on the assumption that these ecosystems have a similar structure to the habitats in the study area, the aforementioned mean value was used to select the most appropriate biomass estimation equation (or equations) for the habitat.

## 3. Results

The mean value of the BEF, as calculated for holm oak (*Quercus ilex* L.) ecosystems in Catalonia, was used to predict the biomass for various values of standing tree volume (Table 3). These values were compared with the predictions from the seven equations in Figure 5, in order to select the most suitable one. The table below presents the BEF predictions, and the predicted values as derived from the seven published equations.

Volume m <sup>3</sup> ha <sup>-1</sup>	M BEF	$M_3$ Mg ha $^{-1}$	$M_4$ Mg ha $^{-1}$	$M_5 \ { m Mg} \ { m ha}^{-1}$	$M_6 \ { m Mg} \ { m ha}^{-1}$	$M_7$ Mg ha $^{-1}$	$M_8$ Mg ha $^{-1}$	$M_9$ Mg ha $^{-1}$
10	12.80	20.07	38.40	36.95	32.11	18.74	26.19	27.47
20	25.60	38.26	53.96	49.42	39.04	23.83	37.39	37.22
30	38.40	56.44	69.51	61.89	45.97	28.92	48.59	46.96
40	51.20	74.63	85.07	74.36	52.91	34.02	59.79	56.71
50	64.00	92.81	100.62	86.83	59.84	39.11	70.99	66.45
60	76.80	110.99	116.18	99.29	66.78	44.20	82.18	76.19
70	89.60	129.18	131.73	111.76	73.71	49.29	93.38	85.94
80	102.40	147.36	147.29	124.23	80.64	54.38	104.58	95.68
90	115.20	165.55	162.84	136.70	87.58	59.47	115.78	105.43
100	128.00	183.73	178.40	149.17	94.51	64.56	126.98	115.17
110	140.80	201.91	193.95	161.64	101.45	69.65	138.18	124.91
120	153.60	220.10	209.51	174.11	108.38	74.74	149.38	134.66
130	166.40	238.28	225.06	186.58	115.31	79.83	160.58	144.40
140	179.20	256.47	240.62	199.05	122.25	84.93	171.78	154.15
150	192.00	274.65	256.17	211.52	129.18	90.02	182.98	163.89

**Table 3.** AGB in Mg ha<sup>-1</sup> as calculated by BEF and the seven published equations in Figure 5. The index number in the header of each column corresponds to the equation in Table 1. Numbers in bold indicate values close to biomass estimated from BEF.

Equations (8) and (9) provide a more accurate approximation of the AGB per unit area compared to the values predicted through the BEF. Therefore, they were selected for the development of the generalized allometric equation. The process of developing the generalized equation is described by several studies [24,51,52]. For *Quercus ilex* L. trees in the study area, this equation was formulated as follows:

$$M = 0.2278D^{2.1335} \tag{11}$$

and was applied to the diameter (D) distributions of the sampling plots, as recorded in the field. The AGB (trunk + branches + foliage) was estimated for each tree in the study area using Equation (11). Subsequently, the total biomass was calculated for each SP and then it was converted to "per hectare" ( $ha^{-1}$ ) figures (Table 4).

**Table 4.** AGB (M) in Mg ha<sup>-1</sup> as calculated by the general Equation (11) for the 43 SPs.

SP	$M$ (Mg ha $^{-1}$ )	SP	$M$ (Mg ha $^{-1}$ )
1	39.94	23	45.25
2	45.25	24	34.36
3	34.36	25	50.47
4	31.20	26	182.46
5	36.66	27	97.75
6	42.41	28	70.78
7	36.14	29	84.92
8	22.01	30	22.4
9	41.58	31	129
10	21.92	32	69.68
11	31.86	33	205.27
12	16.1	34	136.5
13	34.09	35	178.24
14	28.02	36	178.25
15	36.26	37	30.52
16	34.41	38	36.65
17	39.63	39	48.09
18	21.86	40	40.64
19	36.95	41	57.67
20	29.66	42	148.91
21	38.81	43	75.31
22	39.94		

The range of AGB per hectare for the 43 SPs ranged from 16.10 to 205.27 Mg ha<sup>-1</sup> (mean 61.91 Mg ha<sup>-1</sup>). For the estimation of the total biomass in habitat 9340, two different approaches were applied. For the implementation of the first method, the study area was divided into three geographical regions. The average biomass of the SPs that were closest to each region was used to estimate the above-ground organic matter in areas of the habitat. In the second approach, the estimation for the entire area was based on spatial interpolation using the geographic information system QGIS [60].

# 3.1. Estimation Based on the Mean Average (First Approach)

For the broader area of Skiti, Agiokampos, Polydendri, and Rakopotamos, the mean of the SPs shown in Figure 6 was used, which equals 29.75 Mg ha<sup>-1</sup>. The habitat in this area covers an area of 205.03 ha, so the above-ground biomass was estimated at 6100 Mg. For areas of the habitat located in the locations of Sklithro, Kamari, and up to Veneto (Figure 7), the mean of the biomass values from SPs 25–36 and 41–43 was used, amounting to 112.51 Mg ha<sup>-1</sup>. The habitat covers an area of approximately 978 ha in this region, thus the dry AGB is equal to 110,034 Mg. For areas of the habitat located from Pouri to Kalamaki (Figure 8), the average of the AGB from SPs 1–6 and 40 was calculated and found to be 38.64 Mg ha<sup>-1</sup>. In these regions, the habitat covers an area of 2062.58 ha, so it was estimated to bear a biomass equal to 79,698 Mg. Finally, the total biomass of habitat 9340, across the entire study area, amounts to 198,832 Mg.



**Figure 6.** Map depicting the SPs and segments of habitat 9340 (highlighted in blue), in the broader region of Skiti, Agiokampos, Polydendri, and Rakopotamos.

Legend

0

Sampling plots

Habitat 9340



**Figure 7.** Map depicting the SPs and segments of habitat 9340 (highlighted in blue), in the broader region of Sklithro, Kamari, and Veneto.



**Figure 8.** Map depicting the SPs and segments of habitat 9340 (highlighted in blue), in the broader region of Pouri, Zagora, Tsagarada, and Kalamaki.

## 3.2. Estimation Based on Spatial Interpolation (Second Approach)

The method that was used for the estimation based on spatial interpolation is the inverse distance weighted (IDW) interpolation method. Kriging is best suited to well-distributed data, with no discontinuities [61]. In the present study, the SPs were not distributed in all the areas. In the IDW method, the weights are not affected by the spatial arrangement of the samples [61], therefore the IDW was applied.

The biomass values from the 43 SPs were used for the point-based spatial interpolation process. The fundamental principle of the IDW method is that points neighboring the sampling units have a greater influence compared to points that are more distant from them. Figure 9 presents a map of the wider study area, with the results of the spatial interpolation depicted using shades of brown.



**Figure 9.** Map of biomass estimation using spatial interpolation based on the data from the SPs, for areas of habitat 9340 (official boundaries of the habitat).

The interpolation map consists of pixels, each covering an area of  $148 \times 148$  m, and each pixel carries a specific biomass value in ha<sup>-1</sup> (as obtained using the IDW method). For each area of habitat 9340, the average biomass in ha<sup>-1</sup> was calculated from all the pixels within it. Subsequently, the mean value was multiplied by the corresponding area to calculate the AGB. Finally, by summing up all the areas, the total biomass for habitat 9340 was estimated to be 183,505 Mg. As observed, the two methods for estimating the AGB differ by a minimal percentage, approximately 7%. Therefore, we consider their mean value (i.e., 191,168 Mg) to objectively represent the total dry biomass of habitat 9340 in the study area.

# 3.3. Estimation of Carbon Storage in Habitat 9340

To estimate the carbon content of the dry organic matter from *Quercus ilex* L. trees, the value of 47.2% was used, which was derived from a field study conducted in Catalonian Mediterranean ecosystems (Sabaté et al. [39]). Following the guidelines of the IPCC [3] and due to a lack of relevant data, this percentage was utilized to estimate the carbon storage in the studied forest ecosystems. Thus, the carbon storage in the estimated standing dry biomass in habitat 9340 amounts to 83,725.25 Mg, calculated as 0.472 times 191,168 Mg. Finally, to calculate the sequestered  $CO_2$  in the study area, carbon storage was multiplied by the ratio 44/12, resulting in a final estimate of 306,992.58 Mg.

## 3.4. Estimation of Biomass, Carbon Storage, and CO<sub>2</sub> in Quercus Ilex Communities

In the previous sections, the estimation of biomass, carbon storage, and sequestered  $CO_2$  was based on the habitat 9340 areas of the site, as mapped in the context of the project "Monitoring and assessment of the conservation status of habitat types of Community interest in Greece" (~3245 ha). Nevertheless, using satellite imagery, photointerpretation, and ground observations, the authors estimated that the *Quercus ilex* communities in the GR1420004 and GR1430001 areas in the Natura 2000 network cover an area of approximately 5230 ha (Figure 10). The total biomass in these communities was estimated at 261,809 Mg (using the IDW method), the carbon storage was calculated to be 123,573 Mg, and the amount of sequestered  $CO_2$  was found to be 453,104 Mg. The results of the above analyses are summarized in Table 5.

**Table 5.** Summarized estimates of AGB, carbon storage, and CO<sub>2</sub> for Quercus ilex in habitat 9340 and in study area.

	Q Mean Value (1st Approach)	). <i>ilex</i> in Habitat 9340 IDW (2nd Approach)	Mean Value	<i>Q. ilex</i> in Total Area IDW
Dry biomass (Mg) Carbon storage (Mg) Sequestered CO <sub>2</sub> (Mg)	198.832	183.505	191.168 83.725 306.992	261.809 123.573 453.104



**Figure 10.** Map of biomass estimation using spatial interpolation of biomass data from 43 SPs, for the entire area of the habitat.

# 4. Discussion

Climate change has the potential to profoundly impact the distribution, the structure, and functions of forest ecosystems in Greece. This carries significant ecological, economic, and social implications, highlighting the urgent need for appropriate mitigation strategies [62]. Our findings suggest that *Quercus ilex* forests could play a significant role towards climate change mitigation with proper management, as the estimated CO<sub>2</sub> sequestration was 306,992 Mg. Nevertheless, it should be noted that the main factors influencing organic matter production in forest ecosystems vary depending on the soil conditions, microclimatic characteristics, stand age, and management methods, etc. It is immediately understood that the application of a generalized allometric equation for estimating above-ground biomass introduces errors, which can only be determined by comparing the predicted values with field data. Therefore, due to the lack of empirical biomass data, the comparison can be made against published mean values in forest ecosystems.

The range of the AGB per hectare for the investigated SPs varied from 16.10 to  $205.27 \text{ Mg ha}^{-1}$  (mean value of 61.91 Mg ha<sup>-1</sup>), falling within the boundaries of similar studies. More specifically, in Montpellier, France the total AGB (trunk, branches, foliage) was estimated at 71.4 Mg ha<sup>-1</sup>, following the application of appropriate allometric equations developed for the study area [38], while the estimated AGB of the evergreen oaks

in two experimental stands in the Moroccan Middle Atlas amounted to 96 Mg ha<sup>-1</sup> and 86.4 Mg ha<sup>-1</sup> [41]. Therefore, we consider that the generalized equation yielded results that are comparable to the biomass values of similar ecosystems, thus reducing the likelihood of underestimated amounts of above-ground organic matter for habitat 9340.

Focusing on the merchantable biomass for *Q. ilex* L. in Spain, the BEF was found to be  $1.28 \text{ Mg m}^{-3}$  (sd = 0.15), and the mean AGB per unit area was 59.5 Mg ha<sup>-1</sup> (sd = 43.6) [39]. The above parameters were calculated using the allometric equation from the Forest Census of Catalonia [63], which estimates tree AGB based on the diameter at breast height. Additionally, subsequent analyses revealed that the mean carbon content in the woody parts of *Q. ilex* was 47.2% (range 46–48%). It should also be emphasized that the first and only attempt to estimate forest biomass at the Greek national level was conducted for the purposes of the VALOREN project. According to the data analysis, the biomass of Greek forests was estimated at 114 × 106 Mg of dry matter, of which 47% was found in the bark, leaves, branches, roots, and stumps, while the remaining 53% was obtained during logging operations [64].

## 5. Conclusions

This study has analyzed the AGB, carbon storage, and sequestrated  $CO_2$  in habitat 9340 for *Quercus ilex* L. Despite the lack of destructively sampled biomass specimens, the amalgamation of robust statistical techniques with published empirical values provided a solid framework to estimate assimilated  $CO_2$ . Geospatial techniques were also used to derive unbiased extrapolated biomass/carbon values for the study area.

This research has revealed that *Q. ilex* L. plays an integral role in carbon storage, as it was estimated to sequester 306.992 Mg of CO<sub>2</sub>. The results suggest that *Q. ilex* L., a widely spread species in the Mediterranean region, which is found as a shrub in coastal maquis and as a tree in evergreen and mixed Mediterranean forests, could play a significant role in regulating the carbon balance, thereby contributing to the reduction in the atmospheric CO<sub>2</sub> concentration in the Mediterranean basin.

The information acquired in the current research enhances the comprehension regarding the impact of *Q. ilex* L. on carbon storage within the Mediterranean region. A prolonged monitoring of the examined stands, along with the investigation of additional stands in other similar areas of Greece, and in the broader Mediterranean region, could prove beneficial for establishing a Mediterranean database focused on the impact of this type of forest on carbon storage.

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