# Full Length Research paper

# Above-ground tree biomass equations and nutrient pools for a paraclimax chestnut stand and for a climax oak stand in the Sierra de Francia Mountains, Salamanca, Spain

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Above-ground tree biomass and total amounts of nutrients in biomass were estimated at two plots: the first in a *Castanea sativa* Mill. paraclimax coppice and the second in a *Quercus pyrenaica* (Wild.) climax forest, both of them located in the south of the Salamanca (Spain), by harvesting 10 trees at each plot. In order to obtain the best fit possible, for these analyses, four regression equations were applied: linear, logarithmic, potential and exponential. In the chestnut coppice, the best fits were obtained using the exponential regression, while for the oak plot the best fit was obtained with the linear regression. A parametric analysis was also performed for each fraction of the tree biomass between both plots. This revealed that, total biomass and that of the trunks, branches and leaves at the chestnut coppice were significantly greater than those found at the oak plot. At the chestnut coppice for the nutrients (except carbon) significant differences were observed between leaves, branches and trunks, the highest concentrations being found in the leaves, followed by the branches and the trunks. This order was also observed at the oak plot, with the exception of calcium, which did not reveal significant differences between the branches and the trunks.

**Key words:** Above-ground biomass, allometric method, nutrient storage, forest ecosystems, *Castanea sativa*, *Quercus pyrenaica*.

### INTRODUCTION

Currently, the aims of studies addressing biomass are to elucidate energy and nutrient cycles. Such studies are also used to observe the effect of the vegetation on the global CO<sub>2</sub> cycle. Some CO<sub>2</sub> models include the estimation of biomass as the volume, its components, or some related parameters in order to establish the flows of gas between the vegetation, soil, and the atmosphere. Studies of biomass have been carried out in tropical, temperate, and Mediterranean forests and in semi-arid areas (Rapp et al., 1999; Santa Regina and Tarazona, 2000;

2001; Návar et al., 2002; Zianis and Mencuccini, 2003; Nadezhdina et al., 2004; Rubilar et al., 2005; Segura, 2005).

Forest biomass can be calculated using both indirect and direct methods. The former are usually used when tree dimensions are too big, as for example that occurs with many tropical species. In this case, the dimensions of the tree are estimated and the volume of the trunk plus the longest branches is calculated using the formulas of Smalian and Huber (Loetsch et al., 1973). Direct methods, also known as destructive methods, consist of felling trees to determine biomass according to the weight of each of their components: roots, trunk, branches and leaves (Parresol, 1999). Recent studies (Zianis et al., 2005) have shown that, these methods tend to afford

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| Stand  | C. sativa coppice (CC)   | Q. pyrenaica (Oak)         |
|--|--------------------------|----------------------------|
| Altitude (m.a.s.l.)                                      | 1015                     | 950                        |
| Soil type  | Umbric regosol           | Umbric leptosol            |
| L.A.I. (m <sup>2</sup> m <sup>-2</sup> ) leaf area index | 2.9                      | 2.5                        |
| Mean P (mm) (Annual rainfall)                            | 1590                     | 1530                       |
| Mean annual temperature ( º C)                           | 10.8                     | 11.1                       |
| Size of trees (DBH cm                                    | 5.35-19.3                | 5.2-23.6                   |
| Tree age (years)   | 70                       | 75                         |
| Tree height (m)  | $15.3 \pm 1.3$ °         | $12.2 \pm 1.0^{\ b}$       |
| Diameter at breast height (cm)                           | $12.90 \pm 1.7^{ b}$     | 11.60 $\pm$ 1.5 $^{\rm b}$ |
| Stand density (trees ha <sup>-1</sup> )                  | 1 892 ± 100 <sup>b</sup> | $2960\pm125^{\mathrm{c}}$  |
| Basal area (m² ha <sup>-1</sup> )                        | $28.40\pm8^{\circ}$      | $26.50\pm7^{\circ}$        |

**Table 1.** General characteristics of stands selected in the area studied. Confidence intervals p = 0.05. For each site, mean values in the same column followed by different letters are significantly different.

biased values of the weights of the different fractions such that, when a high degree of precision in the estimations is required, it is necessary to employ more specific methods such as "randomized branch sampling" or "important sampling" (Valentine et al., 1984).

Once the weights of the different fractions have been estimated, it is necessary to fit a mathematical model that will relate the weights of the dry biomass to one or more representative variables of the trees, such as: DBH and height. Usually, allometric equations are employed to predict the individual biomass of each tree and the results of these equations are then pooled to obtain the total biomass per area (Parresol, 1999; Socha and Wezyk, 2007; Repola, 2008; 2009; Basuki et al, 2009; Kaonga and Bayliss-Smit, 2010). These equations are developed from tree characteristics that are easy to measure, such as diameter at breast height (DBH), basal diameter (D), height, or a combination of all of them (Parresol, 1999). However, these regression equations may not be so useful when attempts are made to extrapolate them to larger areas (McWilliam et al., 1993; Bollandsas et al., 2009; Liu and Westman, 2009; Durkaya et al., 2010a; 2010b). In the large number of studies addressing tree biomass, many equations have been developed for different species and under different environmental conditions, because biomass values vary with species, age, soil quality and climate (Madgwick and Satoo, 1975).

The nutrient concentration in the leaves of plants provides information about several aspects of plant ecophysiology, such as the photosynthesis and respiration ratio (Reich et al., 1998), growth capacity (Cornelissen et al., 1997), nutrient use efficiency (Chapin, 1980; Aerts and Chapin, 2000), and nutrient limitations (Aerts and Chapin, 2000). Furthermore, the nutrient concentration in leaves affects processes such as leaf litter decomposition and mineralization (Melillo et al., 1982) and damage caused by herbivory and pathogens (Nordin et al., 1998). The main aim of the present work was to approach certain structural characteristics (age, height

and DBH of the trees, and stand density in chestnut stands developed in an area of climax oak. To do so, we report on the regression equations employed for estimating the total aboveground biomass, trunk, branches and leaves in the forests examined. The nutrient distribution and accumulation in different components are also determined.

#### **MATERIALS AND METHODS**

# Field site and experimental set-up

The research was carried out in the Sierra de Francia area, in the province of Salamanca (Spain). Two permanent untilled and unfertilized plots were chosen: the first is *Castanea sativa* (Mill.) paraclimax coppice, and the second is *Quercus pyrenaica* (Wild.) climax forest. The densities of the trees were (tree ha<sup>-1</sup>): 1892 and 2960 for chestnut and oak, respectively. General characteristics of the stands studied are shown in Table 1.

The study area is dominated by granite substrates with generally acidic soil pH and with some limestone intercalations. The soil type varies with depth and among plots. It is classified as umbric regosol in coppice stand and umbric leptosol in oak and orchard plots (FAO., 1989). The zone harbours enclaves of the vegetation typical of the EuroSiberian Region, with the presence of taxa such as *Ilex aquifolium L., Aconitum napellus* ssp., *Castellanum L., Actaea spicata L., Monotropa hypopitys L., Atropa belladona L., Hypericum montanum L., Neottia nidus-avis* Rich., *Paris quadrifólia L., Corylus avellana L.*, etc.

The mean annual temperature is around 10.0°C and in the present study there were only small differences observed among the values obtained for the four years of the study period (2001 - 2004), that is, possibly due to the close location, of the two plots. Mean maximum monthly temperatures were recorded in July and August, with values ranging between 20 and 22°C. The minimum temperatures were recorded in January, the lowest of these being - 1.25°C at the oak stand in January 2003 (Salazar, 2008).

In order to establish differences in the aboveground biomass between the chestnut ecosystem and the climax forest vegetation provided by *Q. pyrenaica*, the aboveground biomass was estimated at both sites. First, the diameter at breast height (DBH) of all the trees was estimated on each plot and grouped by diametric classes. In order to obtain the most accurate estimation of biomass.

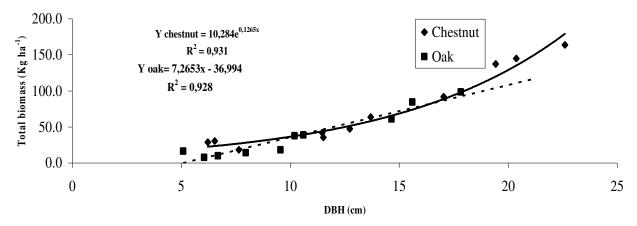


Figure 1. Correlation between total above-ground biomass and DBH for the two species studied.

the most widely used method was employed (Satto and Madgwick, 1982). To accomplish this, allometric relationships between biomass and DBH were used. Ten trees were felled on each site. The felled trees were divided into segments according to their heights (0 - 1.30, 1.30 - 3.0, 3.0 - 5.0, 5.0 - 7.0, 9.0 - 11.0 m and so on, successively) and the leaves, branches, and trunks were separated from each of these segments and weighed in the field.

#### Laboratory methods

Sub-samples of these segments were taken to the laboratory where they were dried in a MEMMERT drying, oven at 80°C, to determine water content and for later analysis (Santa-Regina, 2000). The elements determined in the plant samples were total carbon, total nitrogen, phosphorus, calcium, magnesium and potassium. The analytical methods used are described below: (i) Organic carbon was determined in dry mode with a WÖSTHOFF CARMHOGRAPH 12 (Salazar, 2008).

(ii) Total N was determined with a 3 BRAN LUEBBE Autoanalyzer. Organic nitrogen was mineralized by wet digestion; to accomplish this, 0.1 g of ground homogenized sample was weighed on a Sartorius precision balance and concentrated sulphuric acid plus a small amount of K<sub>2</sub>SO<sub>4</sub>/Se/ Cu SO<sub>4</sub> catalyst was added (Santa-Regina and Tarazona, 2000).

(iii) Phosphorus was determined by colorimetry using the vanadate-molybdate yellow method (Chapman and Pratt, 1979). (iv) Calcium and magnesium were determined with atomic absorption spectroscopy, while potassium was determined with flame photometry. All elements were measured on a VARIAN 220 Fast Sequential atomic absorption spectrophotometer (Santa-Regina et al., 2000).

# Statistical methods and calculations

The DBH and the dry weight of the leaves, branches and trunks from each tree felled were used to calculate different regression equations between DBH and leaf, branch, trunk and total biomass. Statistical analyses were performed using the Statistical Package Social Sciences (SPSS). Because our interest lay in comparing the means of different nutrients under the different forest management regimes and for different years, two- and three-way analyses of variance (ANOVA) were carried out. The ANOVA technique is one of the methods most widely used to analyze simultaneous data when several groups are present and it allows one to check whether there are differences among the means of three or more groups. Once it had been determined whether there were

significant differences in the means of the different groups, the *post-hoc* tests of the SPSS were implemented to determine which of those means were different. Particularly, the least significant difference (LSD) test was employed. In order to obtain the best fit possible, for these regression analyses, four regression equations were used: linear, logarithmic, potential and exponential. In the case of chestnut, the best fits were achieved with the exponential regression, while for the oak plot linear regression afforded the best fit (Figures 1, 2, 3 and 4).

# **RESULTS**

#### Above-ground tree biomass

The tree biomass values (Mg ha<sup>-1</sup>) obtained for the two plots are shown in Figure 5. At chestnut, the wood biomass was 96.3 Mg ha<sup>-1</sup>, 77% of the total biomass and 56.0 Mg ha<sup>-1</sup> at oak (75.7%). The branches were the second fraction is importance, with values of 26.0 Mg ha<sup>-1</sup> at chestnut (20.8% of the total) and 15 Mg ha<sup>-1</sup> at oak (20.3%). Finally, the leaves had a biomass of 2.9 Mg ha at chestnut (2.3% of total) and 3.0 Mg ha<sup>-1</sup> at oak (3%). For both chestnut and oak, ANOVA for wood, branches and leaves, showed significant differences between total biomass, trunk, branch and leaf biomasses, with the following order: total biomass > trunks > branches > leaves (Table 2). Likewise, a parametric analysis was carried out for each fraction of the aerial biomass between the two plots. This revealed that total biomass, trunk, branch, and leaf biomasses at the chestnut plot were significantly higher than the oak plot. Despite the differences in the amount of biomass of the different fractions between the two plots studied, the proportions of each fraction with respect to the total biomass remained similar (Table 2).

#### **Nutrient concentrations**

The chemical compositions of the different tree components at chestnut and oak plots are shown in Table 2.

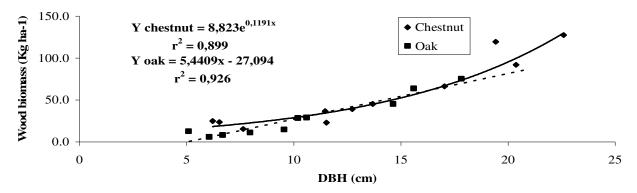


Figure 2. Correlation between wood biomass and DBH for the two species studied.

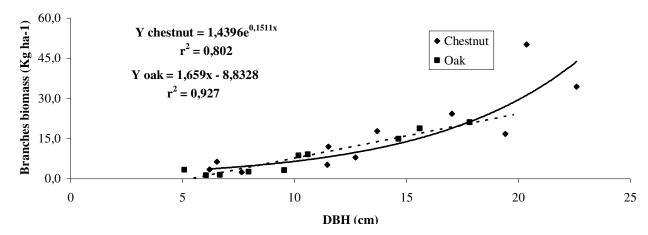


Figure 3. Correlation between branches biomass and DBH for the two species studied.

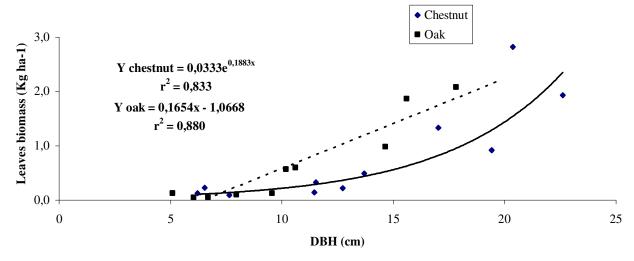
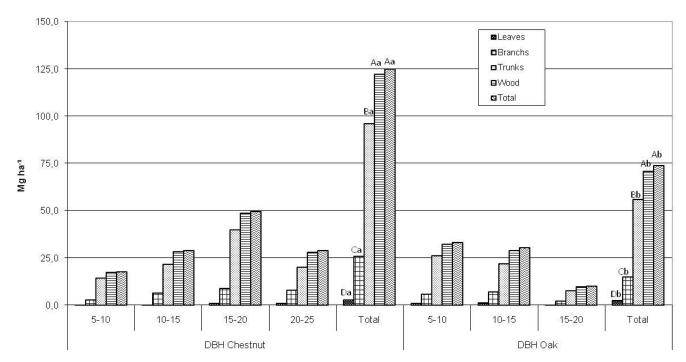


Figure 4. Correlation between leaves biomass and DBH for the two species studied.

According to the found results, there were no significant differences between the two plots for carbon concentrations in the trunks, branches and leaves. However, at

chestnut there were significant differences between the leaves, branches and trunks for the other nutrients, the highest concentrations being observed in the leaves,



**Figure 5.** Above-ground biomass (Mg ha<sup>-1</sup>) of the two plots studied. Different capital letters significantly differences at (p<0.05) according to LSD test, for the different fractions within each plot. Different lower cases significantly differences for each plot within each biomass fraction.

followed by the branches and the trunks. The same concentration order was found at the oak plot, with the exception of calcium and potassium that did not show significant differences between the branches and the leaves. The comparisons between the concentrations of each nutrient in the different considerate parts of the tree are shown in Table 2 for both plots. In the case of the trunks, significant differences were observed for the concentrations of carbon, phosphorus, and potassium. In the branches, significant differences were found for phosphorus, magnesium and calcium. Finally, in the case of the leaves, significant differences were found for phosphorus, magnesium, potassium and calcium.

#### **Nutrient amounts**

The values of mineral mass for chestnut and oak are indicated in Table 3. At both plots, the order of nutrient accumulation in the total biomass was as follows:

In the trunks, significant differences were found for the amount of mineral mass between the two plots studied. At CC the nutrient order was as follows:

C > N > Ca > K > Mg > P while at the Oak plot, the order was: C > N > K > Ca > Mg - P (the Kg ha<sup>-1</sup> of P and Mg were equal).

At both plots, the order of nutrient accumulation in both the branches and leaves was the same. The pattern was as follows:

For the branches: C > C/N > N > Ca > K > Mg > P For the leaves: C > C/N > N > K > Ca > Mg > P.

# **DISCUSSION**

# **Above-ground biomass**

The weights obtained for the leaves, branches, trunks and total biomass were correlated with the DBH using regression analysis. The parameter most widely used as the independent variable is DBH, owing to the ease and precision with which it can be obtained and because it relates the total volume of biomass to functional processes such as transport and the age of the tree (Satto and Madgwick, 1982). Although other models incorporating other variables such as height or basal area may afford good fits, they may often be impractical owing to the difficulty involved in accurately measuring the variables used, especially in dense forests (Segura, 2005). Accordingly, here the DBH was used as the independent variable for the regression analyses. It should be noted that the allometric equations obtained are only valid for a certain interval of the independent variable; extrapolation below or above these ranges could lead to the establishment of marked deviations between the true

**Table 2.** Mean nutrient concentration (mg  $g^{-1}$ )  $\pm$  standard error in leaf and perennial tissues of the different parts of the cut trees in the two plots, chestnut and oak.

| Plot     | Organ    | С              | N              | Р              | Ca             | Mg             | K              |
|----------|----------|----------------|----------------|----------------|----------------|----------------|----------------|
|          | Wood     | 459.3 a (±6.6) | 2.79 c (±0.12) | 0.29 c (±0.01) | 2.66 c (±0.96) | 0.47 c (±0.33) | 0.55 c (±0.05) |
| Chestnut | Branches | 447.3 a (±1.2) | 6.51 b (±0.18) | 0.59 b (±0.04) | 3.83 b (±0.50) | 1.27 b (±0.08) | 2.01 b (±0.09) |
|          | Leaves   | 440.0 a (±8.6) | 14.1 a (±0.56) | 1.11 a (±0.08) | 5.90 a (±0.37) | 3.10 a (±0.21) | 8.96 a (±0.33) |
|          |          |                |                |                |                |                |                |
| Oak      | Wood     | 430.9 a (±5.3) | 3.14 c (±0.25) | 0.40 c (±0.02) | 1.86 c (±0.23) | 0.40 c (±0.04) | 1.97 b (±0.09) |
|          | Branches | 439.2 a (±4.7) | 6.58 b (±0.33) | 0.72 b (±0.04) | 4.30 a (±0.49) | 1.05 b (±0.06) | 1.80 b (±0.21) |
|          | Leaves   | 428.3 a (±7.2) | 14.8 a (±0.25) | 0.87 a (±0.05) | 4.36 a (±0.25) | 1.93 a (±0.11) | 6.5 a (±0.21)  |

**Table 3.** Nutrients immobilized in the above-ground biomass (Kg ha<sup>-1</sup>).

| Plot     | Organs   | С       | N     | C/N     | Р    | Ca    | Mg   | K     |
|----------|----------|---------|-------|---------|------|-------|------|-------|
| Chestnut | Wood     | 44253.8 | 268.8 | 15969.8 | 27.8 | 256.5 | 45.5 | 52.5  |
|          | Branches | 11616.7 | 169,1 | 1834.7  | 15.4 | 99.4  | 33.1 | 52.2  |
|          | Leaves   | 1254.7  | 40.4  | 90.4    | 3.2  | 16.8  | 8.9  | 25.6  |
|          | Total    | 57125.2 | 478.2 | 17895.0 | 46.3 | 372.7 | 87.4 | 130.3 |
| Oak      | Wood     | 24132.8 | 175.8 | 7998.2  | 22.4 | 104.2 | 22.4 | 110.3 |
|          | Branches | 6605.3  | 99.0  | 1119.5  | 10.8 | 64.7  | 15.8 | 27.1  |
|          | Leaves   | 1274.7  | 44.1  | 86.2    | 2.6  | 13.0  | 5.7  | 18.3  |
|          | Total    | 32012.8 | 318.9 | 9203.9  | 35.8 | 181.8 | 43.9 | 155.7 |

and predicted values (Zianis and Mencuccini, 2003).

Extrapolation of the results obtained should be performed with caution since the factors affecting productivity vary considerably in any given forest because it is affected by orientation, soil depth, fertility, the type of substrate, microclimatic characteristics, density, age, type of management, etc. (Leonardi et al., 1992; Rapp et al., 1992). Thus, it is common to obtain incorrect estimations of biomass when the equations or a given zone are extrapolated to other geographic areas (Harding and Grial, 1986; Neyrinck et al., 1998; Wang and Kimmins, 2002: Zianis and Mencuccini, 2003: Zabek and Prescott, 2006). Nevertheless, the extrapolation of results is frequent because of the cost and difficulty of measuring tree biomass (Zabek and Prescott, 2006). Wirth et al. (2004) believe that the regression equations developed in a region can reasonably be used to predict tree biomasses in other places. When it is necessary to estimate forest biomass, there are many predictive equations that allow aerial biomass to be obtained in a non-destructive way. Thus, the literature contains numerous sets of biomass equations for different tree species over a broad range of environmental conditions (Eamus et al., 2000 for Australia; Jenkins et al., 2004; Zianis et al., 2005 for North-America). Most such equations have been developed using trees from a specific site or from places representing only small regions. Therefore, the use of these equations at global

scale is meaningless (Jenkins et al., 2004).

The values obtained for total biomass at chestnut (125.1 Mg ha<sup>-1</sup>) and oak (74 Mg ha<sup>-1</sup>) lie within the range reported by other authors for both species in different geographic locations. Thus, in chestnut coppices devoted to wood production in Spain, Santa Regina (2000) reported values of 120.4 Mg ha<sup>-1</sup>; in France, Santa Regina et al. (2000), Ranger et al. (1990) and Ranger and Colin-Belgrand (1996) established values of 153.3, 120 and 120 Mg ha<sup>-1</sup>; in Italy, values of 107 (La Marca, 1984) and 108 Mg ha<sup>-1</sup> (Cutini, 2000) have been reported, and in Slovakia Tokarár and Krekulova (2004) found biomass values of between 95.1 and 174. 2 Mg ha<sup>-1</sup>. Likewise, the values obtained in this work for the oak plot were in the 64.5 – 131.8 Mg ha<sup>-1</sup> range reported by other authors (Rapp et al., 1999).

# **Nutrient concentrations and mineralomass**

Knowledge of the distribution of nutrients in the different sections making up the above-ground biomass is of great importance for making realistic predictions about the export of nutrients under different forest management systems (Augusto et al., 2000).

According to the results it is clear that the highest nutrient concentrations are present in the leaves. Similar results have been reported by authors such as Rapp et al. (1999), Santa Regina et al. (2000), Caldeira et al. (2002) and Frangi et al. (2005). Leaf nutrient concentrations are affected by different factors, such as the age of the tree, the characteristics of the site, season of the year, etc. (Van den Driessche, 1984).

The amount of nutrients accumulated in leaves was quantitatively lower than in branches and trunks because leaf biomass only represented 2.9 and 3.0% of the total biomass at the chestnut and oak plots, respectively. However, from the qualitative point of view, the amount of leaf nutrients is of great importance because the leaves of deciduous species are subject to annual shedding cycles, with which the leaf biomass returns to the soil (Gallego et al., 1994). Accordingly, this value varies considerably between sites, with a mean storage of 25% for nitrogen and 15% for phosphorus and potassium. This distribution of nutrients has important implications for forest management (Jokela et al., 1981; Saur et al., 1992). However, the distribution of nutrients in trees is strongly related to the biological activity of the different tree compartments, mainly to the physiological activity of the leaves. (Santa Regina, 2000).

### **Conclusions**

The DBH is a strong predictor of biomass for practical purposes, using the allometric equations. As a variable, height does not improve the results that much and it represents an additional hindrance in compiling the data. Better predictions were obtained by pooling the data area sets from the two sites studied than doing it individually for each one. A parametric analysis revealed that total biomass, of the trunk, branch and the leaf biomasses at CC were significantly higher than the Oak plot. Despite the differences in the amount of biomass of the different fractions between the two plots studied, the proportions of each fraction with respect to the total remained similar. At the chestnut coppice, significant differences were observed in nutrient concentrations and content between leaves, branches and trunks, except for the carbon. The highest concentrations were found in the leaves, followed by the branches and the trunks. The same order of concentration was observed at the oak plot, with the exception of calcium, which did not reveal significant differences between the branches and the trunks.

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