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*Full Length Research Paper*

# Assessment of essential oil yield of *Artemisia herba-alba* cultivated in Tunisian arid zone

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**The main objective of this study is to estimate the essential oil (EO) yield of *Artemisia herba-alba* through biomass prediction. EO yield dependent in both the biomass production and its EO content, was determined from the recovered biomass after harvest of the upper half of plant tuft. The bio-volume parameter of twenty cultivated individuals of *A. herba-alba* were estimated four times during four years from height and diameters measurements. In the first harvest, after one year of establishment, several individuals appeared more productive than others. Thus, the canopy diameters of the plant tuft and the bio-volume recorded little significant relationship with the EO yield ($R^2$ = 0.47 and 0.57, respectively). Harvesting the upper half of plant tuft, the spherical volume model developed by the regenerated material of the same individuals resulted in a high correlation between the EO yield and both the mean diameter ($R^2$ = 0.61 to 0.62) and the bio-volume ($R^2$ = 0.66 to 0.83).**

**Key words:** *Artemisia herba-alba*, essential oil, biomass prediction, arid zone, Tunisia.

## INTRODUCTION

Establishment of mathematical equations to evaluate the aerial biomass of plant species is very important in rangeland management (Aboal et al., 2005; Salis et al., 2006; Xu et al., 2006; Abdelkader et al., 2007, 2008). Various approaches were applied with different plant species and have contributed to the establishment of many mathematical models with the aim to predict plant biomass production (Pilli et al., 2006; Areval et al., 2007; Abdelkader et al., 2007, 2008). Different volume parameters of plant species were retained for measurements to estimate biomass production of shrubs and fodder species (Bartelink, 1996; Sebei et al., 2004; Pilli et al., 2006; Abdelkader et al., 2007, 2008). Crown diameters and plant height were the commonly measured parameters for characterizing the biomass of shrubs.

*Artemisia herba-alba* Asso (Asteraceae) is a medicinal and aromatic shrub, growing wild in arid areas of the Mediterranean basin. In Tunisia, this chamephytic species is found from the mountains around Jebel Oust (Fahs) to the South of the country (Mighri et al., 2009a). This plant is widely used in traditional medicine in treating bronchitis, diarrhea and neuralgias (Akrout, 2004). For instance, it is one of the most frequently used plants to treat diabetes and hypertension in the Errachidia province of Morocco (Tahraoui et al., 2007). The essential oil (EO) of this species was known by its therapeutic disinfectant, anthelminthic and antispasmodic virtues (Hatimi et al., 2001; Houmani et al., 2004; Mighri et al., 2010). To fulfill the current and future increasing demand, cultivation systems that produce a greater amount of EO with good quality are required.

A previous study concerning this aromatic plant cultivated in the arid zone of southern Tunisian (without using fertilizers and with irrigation kept at a minimum), showed that the highest EO yield was obtained from the upper half of plant tuft at the flowering stage with almost the same chemical composition as the oil isolated from wild growing plants (Mighri et al., 2009b). Although, yearly individual biomass production varied, the EO content distilled from the dried biomass remained practically constant.

Continuing our investigation, this study aimed to assess the EO yield through biomass prediction. It is expected, in the future, that *A. herba-alba* be cultured by local farmers and/or EO producers in arid zone of Southern Tunisia. Establishment of mathematical models between the available aerial biomass used for EO distillation and volume parameters (Mean diameter and bio-volume) of plant tufts, could be of a great interest in evaluating the

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**Table 1.** Principal characteristics of individual plants of *Artemsia herba-alba* harvested four times during the experiment (2003 to 2006).

| Year | Height (cm) | | | Mean diameter (cm) | | | Bio-volume ($dm^3$) | | | Dry biomass (g) | | | Essential oil yield | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Min. | Max. | Mean | Min. | Max. | Mean | Min. | Max. | Mean | Min. | Max. | Mean | mL.100 $g^{-1}$ | mL.$m^{-2}$.$yr^{-1}$ |
| 2003 | 38 | 58 | $50^{bc}$ | 82 | 118 | $99^{b}$ | 176 | 423 | $261^{c}$ | 253 | 1.14 | $557^{b}$ | $2.16^{a}$ | $3.26^{ab}$ |
| 2004 | 50 | 67 | $56^{b}$ | 107 | 165 | $133^{a}$ | 300 | 915 | $534^{b}$ | 545 | 1.92 | $1.05^{a}$ | $1.45^{c}$ | $2.89^{b}$ |
| 2005 | 59 | 74 | $66^{a}$ | 125 | 165 | $143^{a}$ | 491 | 997 | $711^{a}$ | 622 | 1.76 | $1.04^{a}$ | $1.85^{b}$ | $3.74^{a}$ |
| 2006 | 46 | 66 | $57^{b}$ | 91 | 154 | $126^{a}$ | 199 | 649 | $473^{b}$ | 227 | 937 | $581^{b}$ | $1.70^{b}$ | $2.59^{b}$ |

Values within a column followed by different letters are significantly different ($P < 0.05$).

*A. herba-alba* EO yields.

## MATERIALS AND METHODS

### Plant material

Field experiments were conducted during four years (2003 to 2006) at the experimental station of the Arid Lands Institute (Tunisia) (33°30'N and 10°40'E). This site has arid-type climate. The mean annual rainfall recorded from 1992 to 2001, is 155 ± 46 mm, with most rainfall falling in the autumn and winter seasons. The average monthly temperature is 21°C, the highest (35°C) being in August and the lowest (5°C) in January. Seedlings of *A. herba-alba* were transplanted in November 2002 at 150 cm row-to-row and plant-to-plant spacing without using fertilizers. Irrigation was kept at a minimum, only in order to prevent the wilting of plants. Measurements are about sixteen individuals of *A. herba-alba* randomly sampled from the experimental field. The plant height (H) and the mean canopy diameter (Md) (as the average diameter of the longest diameter ($d_1$) and the greatest diameter ($d_2$) taken at the widest part of the tuft perpendicular to $d_1$) were measured. Subsequently, each individual plant was harvested at the upper half of *A. herba-alba* tuft. Fresh biomass was dried in shade at ambient temperature until the weight is constant and then separately weighed. After removing stems which does not contain volatiles, EO was extracted from pooled dry matter (DM) constituted by leaves and flowers (100 g each mixed with 600 mL of distilled water) in 2 L capacity round bottom flask in modified Clevenger apparatus for 4 h at each harvest. The content of oil (v/w vs DM) was expressed as mean values of three experiments, which was used to calculate the EO yield (mL. $m^{-2}$. $year^{-1}$).

The bio-volume (BV) corresponding to the volume occupied by the portion of the upper half of *A. herba-alba* tuft was simulated to a spheroid model. This approach was applied with other species such as *Agropyron desertorum* (Johnson et al., 1988), *Atriplex canascens* (Thomson et al., 1998) and *Cenchrus ciliaris* (Abdelkader et al., 2007). The volume model attributed to individuals of *A. herba-alba* was a spheroid:

$$[BV = 2/3\ \pi\ (1/2\ (d_1/2 + d_2/2)^2\ H/2)]$$

After one year of establishment, in the first harvest applied in November 2003 (full blossom), the EO yield for each individual was related to the Md and the BV. These relationships were tested three times in November during three years (2004, 2005 and 2006) with the regenerated material of the same individuals.

### Statistical analysis

The data on plant height, mean diameter, bio-volume, biomass production and EO yield were subjected to analysis of variance (ANOVA). Mean values were considered significantly different when $p<0.05$. Different regressions were generated between the EO yields and both the Md and the BV and the best models were retained.

## RESULTS

Concerning the biomass production, harvesting of upper half (level of cutting 50% of the whole plant) of *A. herba-alba* tuft at the flowering stage, lead to maximum biomass quantity in the second and third harvests (2004 and 2005) compared to the first and the fourth harvests (2003 and 2006) (Table 1). After removing stems, the ligneous part of individuals represented 37, 43, 44 and 60% of the totality of the biomass respectively, in November 2003, 2004, 2005 and 2006. The corresponding EO content extracted from the DM (leaves and flowers) varied in an unpredicatable manner. Indeed, it was higher in first harvest, decreased in second harvest to reach minimum and increased in third and fourth harvests (Table 1). The EO yield dependent in both the DM quantity and its EO content was significantly higher in November 2005 compared with others harvests at the same time to higher biomass yield and EO content.

We remember that, the main objective of this study was to found relationships between the EO yield of *A. herba-alba* plant cultivated in Tunisian arid zone and both the mean canopy diameter (Md) and the plant volume (BV). Principal characteristics of volume parameters of individual plants of *A. herba-alba* determined during the experiment are shown in Table 1.

The EO yields of all individuals (2003 to 2006) were related to the BV and Md. As shown in Figure 1 and Table 2. The linear regression model fitted a relatively highly significant relationship between the EO yield and the BV in the first harvest applied in 2003 ($R^2$ = 0.57). This coefficient of determination increase gradually in the three subsequent harvests (2004, 2005 and 2006) ($R^2$ = 0.66, 0.70 and 0.83, respectively).

It seems that the EO yield is well correlated to the BV especially for the renewal harvests. This could be important for estimating the EO yield. Little relationship was found between the EO yield and the Md. This last model fitted a linear regression with a coefficient of determination of 0.47 in the first harvest applied in 2003

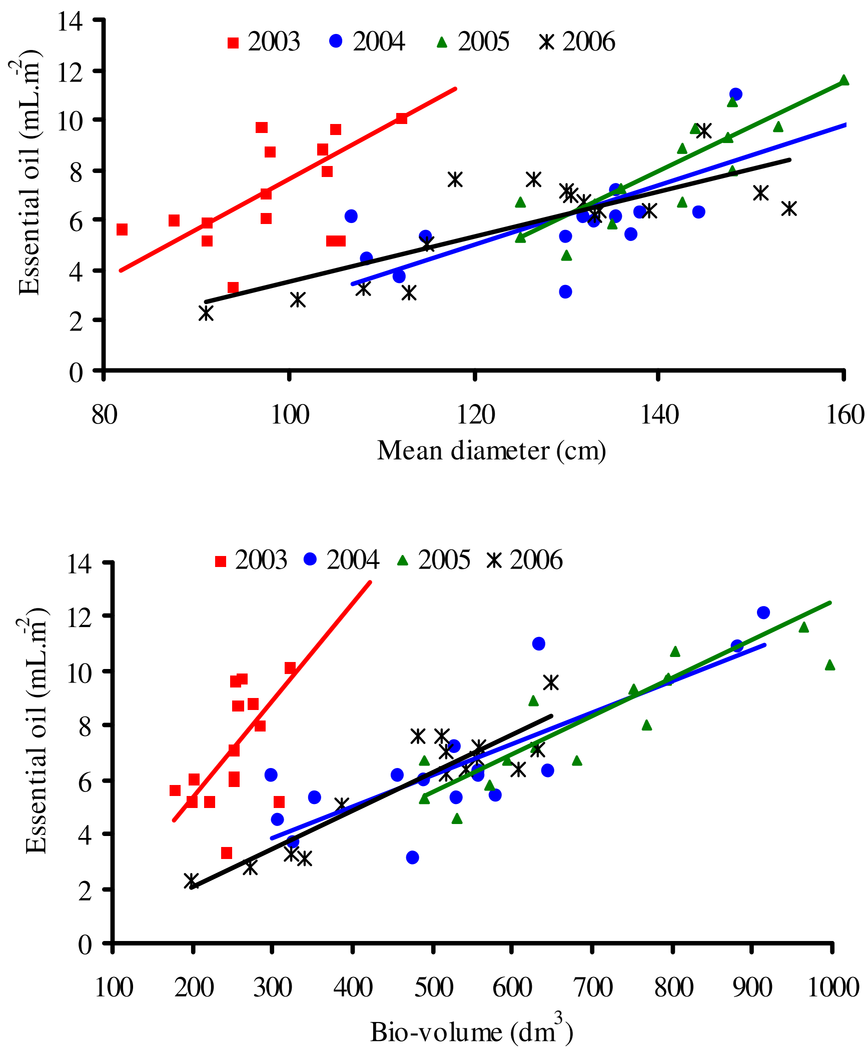


**Figure 1.** Correlations between the essential oil yield and both the bio-volume and the mean diameter of same individual plants of *Artemisia herba-alba* repeated four times in November 2003, 2004, 2005 and 2006.

**Table 2.** Regression models between the essential oil yield (Y) and both bio-volume and the mean diameter (X) of individual plants of *Artemisia herba-alba* harvested four times during the experiment (2003 to 2006). ($R^2$: coefficient of determination).

| Year | (X) | Regression equation | $R^2$ |
|---|---|---|---|
| 2003 | Bio-volume | Y = 0.017 × - 0.78 | 0.57 |
| | Mean diameter | Y = 0.089 × - 5.55 | 0.47 |
| 2004 | Bio-volume | Y = 0.005 × + 0.18 | 0.66 |
| | Mean diameter | Y = 0.052 × - 4.08 | 0.62 |
| 2005 | Bio-volume | Y = 0.006 × - 0.64 | 0.70 |
| | Mean diameter | Y = 0.067 × - 6.00 | 0.61 |
| 2006 | Bio-volume | Y = 0.006 × - 0.33 | 0.83 |
| | Mean diameter | Y = 0.04 × - 2.43 | 0.61 |

(Table 2 and Figure 1). Showing similar trend to the BV, the EO yield appeared well correlated to the Md for the renewal harvests when the coefficient of determination increased and reach a significant value of 0.62.

## DISCUSSION

Our results demonstrate that in an arid climate, harvesting of the upper half of *A. herba-alba* significantly influenced crop growth in respect to Md and BV of the plant tuft and consequently the corresponding quantity of produced biomass and its EO content. Similar results obtained with others aromatic species, shows that the irregularity on biomass production and its EO content might be related to several factors such the various chemotypes, the distilled plant part, the season of harvest and the site of harvest (Rajeswara, 1997; Puttanna et al., 2001; Khotari et al., 2004; Baydar and Baydar, 2005; Rohloff et al., 2005). The relative knowledge of the ecology of plant species wild grown or cultivated in the arid zone is still fragmentary. In this study, no clear relationship between the fluctuation of biomass production and changes in water soil availability or temperatures was found during the experiment (2003 to 2006).

After one year of establishment, several individual plants of *A. herba-alba* appeared more productive than others. This irregularity of biomass production is probably attributed to the age of the transplanted plants (Pilli et al., 2006) and their physiological behavior such the trade-off between vegetative and reproductive allocation of resources (M'seddi et al., 2002). The EO yield, depends on both the quantity of produced biomass and its corresponding EO content, appeared consequently less correlated to the BV and Md.

The higher significant quantities of the recovered biomass in November 2004 and 2005 confirms the previous observations carried out in southern Tunisia, which suggested that individuals species of *A. herba-alba* should be deprived of their old biomass to occur again and to be more productive (Le Houerou, 1995; Mighri et al., 2009). After three successive harvests, the highest density of leaves and floral buds recorded in November 2006 from the basal part of the plant tuft explains this reduction of recovered biomass since we sampled the upper half of the plant.

Harvesting each year the same plants three times in November 2004, 2005 and 2006, it appeared that the BV and the Md were well correlated to the EO yield compared to the first harvest applied after one year of establishment. These changes could be explained by the change in morphology of *A. herba-alba* after harvest, which develops its regenerated aerial parts at the same time in diameter and height, keeping with this manner the spherical volume model of the plant tuft. According to volume parameters measurements (Thomson et al., 1998; Abdelkader et al., 2008), the applied methods on *A. herba-alba* showed high significant relationships between different volume parameters and the EO yield, especially, for the renewal harvests of same individual plant. For producers of *A. herba-alba* EO, from the regenerated biomass, the stability of the EO composition is of particular importance (Mighri et al., 2009b). Before harvesting, our results provide a strong foundation for use of morphometric indices as an adjunct to field methods for assessing EO yield through biomass prediction with a relatively high significance level.

In conclusion, according to all results cited previously, the use of the plant volume as a predictor for EO yield of *A. herba-alba* cultivated in Tunisian arid zone seems to be of great interest. Although in this area, significant changes in respect to biomass production and EO yield were observed from year to another, the canopy diameters ($d_1$ and $d_2$) that are the easily measured variables, illustrate relatively little relationship with the EO yield than the bio-volume. It is expected, in the future, that *A. herba-alba* be cultured by local farmers and/or EO producers in arid zone of Southern Tunisia. Based on these volume parameters (H, $d_1$ and $d_2$), EO yield can be estimated with high significant relationships especially for the renewal harvests.

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