

Research Paper

Morphological traits and essential oil yield variation of three *myrtus communis* L. populations: implication for domestication

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Myrtle is an aromatic plant typical of the Mediterranean maquis. It is widely exploited in pharmaceutical, cosmetic, and food industries. In Morocco, the demand on *Myrtus communis* L. was affected negatively in the last few years and its cultivation is becoming more important. However, little information is available about its intraspecific variation and adaptability. The analysis of variance showed low population effect but a high significant variation within population of most morphological traits and essential oil yield. Among all traits studied, fruit and leaf traits and essential oil yield presented more heterogeneity and high to medium heritability. A clinal altitude, precipitation and

longitude of leaf traits and essential oil content variation is showed. Moreover, strong and positive correlation was found between fruit, leaf and some seed traits, and between essential oil yields. Comparing all traits; fruit trait, essential oil yield and their associated traits could be good criteria for selection Moroccan myrtle at the segregating generations in the future plant domestication and breeding programs.

Key words: *Myrtus communis* L., genetic variability, morphological characters, essential oil yield, phenotypic plasticity.

INTRODUCTION

Myrtle (*Myrtus communis* L.) is one of the most important medicinal and aromatic plants (MAP) in Mediterranean region and the Middle East (one of the most important species of the maquis), where it grows wild and is also cultivated (Sabiha *et al.*, 2011; Migliore *et al.*, 2012). It is grown for its ornamental value and aromatic properties, and its usage has great importance even nowadays. Throughout history, the combination of the myrtle flower's attractive appearance, its edible berries, sweet smell and medicinal traits has made the plant very appealing to the people (Stuart, 1994). Nowadays, the myrtle plant and its secondary metabolites are being tested scientifically in

order to feed future research in cosmetic, medicine, food industry and in biological plant protection (Mulas *et al.*, 2002; Sabiha *et al.*, 2011; Wahid, 2013). For example, extracts and some isolated compounds of this plant have been reported to be efficient as antibacterial, antifungal, anti-inflammatory, antioxidant and hypoglycemic agents by some publishable studies (Alem *et al.*, 2008; Sabiha *et al.*, 2011; Cannas *et al.*, 2013; Gonçalves *et al.*, 2013). The liqueur produced from myrtle is now one of the most promising natural foods, it may be obtained by cold infusion of berries or young spring shoots (Satta *et al.*, 2011), the fields of domestic chemistry, cosmetic and

Table 1. Geographical locations and ecological conditions of myrtle natural populations collected in Morocco.

| Regions | Sites | Latitude North | Longitude West | Altitude (m) | P (mm) | Tmax (°C) | Tmin (°C) |
|--------------------------|-------------|----------------|----------------|--------------|--------|-----------|-----------|
| Occidental Rif | Chefchaouen | 35°10' | 5°19' | 600 | 800 | 36 | -8 |
| Oriental Rif | Taounate | 34°43' | 4°39' | 700 | 750 | 40 | 5 |
| Central Oceanic Atlantic | Rabat | 33°54' | 6°50' | 126 | 500 | 30 | 8 |

P is the mean annual precipitation, Tmax is the maximum average temperature of the warmest month, Tmin is the average minimum temperature of the coldest month.

food industry there is an increasing demand for myrtle. Additionally, its therapeutic indications are extended continuously due to the improved research techniques.

The development of product's myrtle market brought to an increase in harvesting from wild plants that caused progressive impoverishment of wild germplasm and decrease in natural sources of supply (Wahid, 2013). For example, in Morocco, this species contributes to a large fraction of the total exports (45%) of the MAP's Essential Oils (EO). However, plant leaves used for the extraction of EO is exploited from spontaneous range of the species. Although the production of EO from spontaneous plants is important, it is often linked to climate fluctuation and area's species regressed. Thus the demand on the Moroccan myrtle was affected negatively in the last few years (Wahid, 2013). In addition, the spontaneous and often abundant harvest of plant leaves is linked to market pressure. Moreover, some troubles in standardization of the final product derived from spontaneous sources of material were evidenced. This is due to the heavy influence of climatic conditions on the amount and quality of wild plants' production (Mulas *et al.*, 2000). Although myrtle is traditionally a collected aromatic and medicinal plant, its cultivation is becoming more important not only in Morocco but also world-wide. Cultivation of myrtle following genetic breeding and intensive crop management models, could solve those problems (Mulas *et al.*, 2002; Satta *et al.*, 2011; Wahid, 2013). Additionally, production of high quality raw material called "Moroccan" can help to maintain the competitiveness on the international market as well. This can be achieved by using selected and agronomically-known propagation material, obtained through selected genotype.

The sustainable supply via selected genotype, in order to respond to industrial needs, is based on genetic diversity/variability (Migliore *et al.*, 2012, Wahid, 2013). In Morocco, genetic variability of morphological traits within wild myrtle populations is not widely evaluated yet, whereas the chemical characteristics have been partly revealed (Farah *et al.*, 2006; Cherrat *et al.*, 2014). The current knowledge about its morphology and chemical traits, variety development and agronomy is neither complete nor conclusive under Moroccan conditions. The knowledge of such aspect is important not only from a botanical point of view or forest resources management,

but also represents a basic step in developing breeding program or for selecting original plant material with improved ornamental/chemical traits. A lot of work remains to be done to define the potential of this natural endangered resource in Morocco. Hence, the aim of this study was to describe the morphological and chemical traits of myrtle natural populations, and identify natural populations that may express interesting traits, also to evaluate the genetic variability of morphological and chemical traits and it's depending on environmental influences, the study also estimated the correlation among morphological and chemical traits and relate these to the performance of each population. Finally the study explores the correlation between the ecological, geographical conditions and the analyzed traits.

MATERIAL AND METHODS

Plant material

The distribution of myrtle in Morocco is discontinuous, covering different biogeographic regions, which has resulted in local adaptation. Fruits and aerial parts were collected systematically in November 2012 from three natural populations of the species distributed in different biogeographic regions: Chefchaouen from Occidental Rif, Taounat from Oriental Rif, and Rabat from Central Oceanic Atlantic. Its geographic locations and ecological conditions are shown in (Table 1). The tested trees per population ranged from 10 to 20 depending on the seed bearing specimens at each population and according to the population size. Trees were chosen randomly with no phenotypical selection, and were at least 20 m away from each other to ensure inclusion of maximum genetic variation and to avoid sampling from related individuals. The sampling transect method was chosen in the case of localized populations along the ravines drain funds from Chefchaouen sites, and the mode plots for populations' Taounate and Rabat.

Then the seeds and aerial parts from individual trees were sampled by number, and bulked by population for the experiments. For this, thirteen genotypes/ individual (tree)/ population were randomly sampled for measurement of morphological and EO traits. A total number of 520 genotypes (13 genotypes x 40 trees) were

Table 2. Morphological traits measured in *Myrtus communis* L. populations from Morocco.

| Traits | Code | Method of measurement |
|-------------------|---------|-----------------------|
| Fruit traits | | |
| Fruit length | FL(cm) | Caliper |
| Fruit width | FWD(cm) | Caliper |
| Fruit weight | FWG(g) | Precision balance |
| Seed traits | | |
| Seed number | SN | Counted |
| Seed length | SL(cm) | Caliper |
| Seed width | SW(cm) | Caliper |
| Total seed weight | TSW(g) | Precision balance |
| Leaf traits | | |
| Leaf length | LL(cm) | Caliper |
| Leaf width | LW(cm) | Caliper |

used for morphological and EO measurement/determination.

Climatic conditions were evaluated by using the data of meteorological stations, near to the analyzed myrtle populations.

Morphological measurement

Nine morphological traits were examined on trees from natural populations. The data were evaluated on 13 representative genotypes/ tree/ population for several morphological traits (Table 2) to assess the genetic variability among and within populations. The morphological traits examined include fruit length (FL, cm), fruit width (FWD, cm), fruit weight (FWG, g), seed number (SN), seed length (SL, cm), seed width (SW, cm), total seed weight (TSW, g), leaf length (LL, cm), and leaf width (LW, cm).

Essential oil extraction

The leaves of the samples were separated from the stems by hand and weighed. A sample of 100 g of fresh leaves was taken from each tree/ population. Then 200 ml of tap water was added to each sample and water distillation was run for 2 h using a Clevenger-type apparatus as illustrated by (Guenthere, 1972). The essential oil rate was measured by the volumetric method (v/w) (Wichtl, 1971). Thus, the essential oil content percent (R_{EOC} , %) for each genotype was determined on volume of essential oil (V_{EO} , ml) by dry weight of leaves (DWL, g), all multiplied to 100; as $R_{EOC} = (V_{EO}/DWL) \times 100$.

Data analysis

The data were analyzed statistically to find the significance

of the results of morphological characters and essential oil content percent of *Myrtus communis* L. and relationship between traits. The assumptions related to analysis of normal distribution and homogeneity of variance was verified using univariate statistic test and standard graphical methods.

Data transformations were not considered necessary to satisfy these assumptions except for essential oil content percent, which data were subjected to the Log (HE+1) transformation before analysis. The following general linear model and the restricted maximum likelihood (REML) method were used to compute variance components with the MIXED procedure:

$$Y_{ij} = \mu + P_i + I(P)_{j(i)} + e_{ij}$$

where Y_{ij} is the observation from the i^{th} population of the j^{th} individual, μ is overall mean of each trait, P_i is the random effect of i^{th} population with variance σ_p^2 and mean 0 ($i = 1- 3$), $I(P)_{j(i)}$ is the random effect of the j^{th} individual nested within the i^{th} population with variance σ_f^2 and mean 0 (j between 10 and 20 individuals), and e_{ij} is random error with variance σ_e^2 and mean 0. Variance components, expressed as coefficient of variation among populations (CV_p) and within population (CV_f), were estimated as:

$$CV_p = \frac{\sigma_p}{\mu} \times 100, \text{ and } CV_f = \frac{\sigma_f}{\mu} \times 100,$$

where μ is overall character mean. In this study we refer to broad sense heritability (H^2), which is defined as

$$\text{(Zheng et al., 2008; Bilir and Betul, 2013): } H^2 = \frac{\sigma_f^2}{\sigma_f^2 + \sigma_e^2}.$$

Correlation between population means for each morphological trait, essential oil content percent, and environmental factors such as altitude, latitude, longitude and precipitation were studied using Pearson's correlation coefficient. All statistical analyses were carried out using SAS® statistical package, version 8 (SAS, 2009).

Table 3. Descriptive analysis of morphological traits and essential oil content of three natural populations of Moroccan *Myrtus communis* L..

| Populations | FL (cm) | FWD (cm) | FWG (g) | SN | SL (cm) | SW (cm) | TSW (g) | LL (cm) | LW (cm) | EO (%) |
|-----------------------------|---------|----------|---------|-------|---------|---------|---------|---------|---------|--------|
| Population from Chefchaouen | | | | | | | | | | |
| Mean | 0.87 | 0.721 | 0.234 | 6.638 | 0.349 | 0.225 | 0.087 | 3.804 | 1.564 | 0.338 |
| Minimum | 0.580 | 0.490 | 0.078 | 1.00 | 0.260 | 0.170 | 0.018 | 2.530 | 0.780 | 0.262 |
| Maximum | 1.21 | 0.95 | 0.462 | 22.00 | 0.44 | 0.670 | 0.640 | 5.610 | 3.100 | 0.415 |
| Std Dev | 0.132 | 0.091 | 0.083 | 3.235 | 0.0389 | 0.049 | 0.058 | 0.796 | 0.507 | 0.044 |
| Population from Taounat | | | | | | | | | | |
| Mean | 0.86 | 0.66 | 0.204 | 5.12 | 0.35 | 0.27 | 0.069 | 3.27 | 1.16 | 0.292 |
| Minimum | 0.58 | 0.48 | 0.085 | 1.00 | 0.26 | 0.157 | 0.02 | 2.55 | 0.80 | 0.258 |
| Maximum | 1.5 | 0.94 | 0.620 | 19.00 | 0.94 | 0.381 | 0.51 | 4.85 | 1.65 | 0.358 |
| Std Dev. | 0.181 | 0.120 | 0.094 | 2.866 | 0.064 | 0.041 | 0.050 | 0.494 | 0.183 | 0.028 |
| Population from Rabat | | | | | | | | | | |
| Mean | 0.85 | 0.63 | 0.195 | 7.91 | 0.33 | 0.22 | 0.078 | 2.83 | 1.13 | 0.306 |
| Minimum | 0.66 | 0.35 | 0.095 | 2.00 | 0.28 | 0.16 | 0.023 | 2.010 | 0.64 | 0.258 |
| Maximum | 1.15 | 1.09 | 0.466 | 20.00 | 0.44 | 0.33 | 0.66 | 3.870 | 1.970 | 0.396 |
| Std Dev | 0.099 | 0.080 | 0.058 | 3.918 | 0.039 | 0.042 | 0.058 | 0.461 | 0.239 | 0.04 |
| Total Mean | 0.867 | 0.672 | 0.211 | 6.554 | 0.346 | 0.238 | 0.078 | 3.301 | 1.286 | 0.312 |

Table 4. Estimate of variance components (σ), coefficient of variation (CV) and broad-sense heritability (H^2) of morphological traits and essential oil content of three natural populations of Moroccan *Myrtus communis* L.

| Traits | $\hat{\sigma}_p^2$ ($P>Z^*$) | $\hat{\sigma}_f^2$ ($P>Z^*$) | $\hat{\sigma}_e^2$ ($P>Z^*$) | CV _p (%) | CV _f (%) | H ² |
|--------|--------------------------------|--------------------------------|--------------------------------|---------------------|---------------------|----------------|
| FL | 0.0000 ($P= 0.987$) | 0.0037 ($P= 0.002$) | 0.0005 ($P< 0.001$) | - | 0.427 | 0.881 |
| FWD | 0.0015 ($P= 0.232$) | 0.0052 ($P< 0.001$) | 0.001 ($P< 0.001$) | 0.220 | 0.726 | 0.98 |
| FWG | 0.0001 ($P= 0.469$) | 0.0001 ($P= 0.300$) | 0.0031 ($P< 0.000$) | 0.047 | 0.047 | 0.031 |
| SN | 1.3840 ($P= 0.240$) | 5.1958 ($P< 0.001$) | 6.6316 ($P< 0.001$) | 21.12 | 79.277 | 0.439 |
| SL | 0.0001 ($P= 0.431$) | 0.0007 ($P\leq 0.001$) | 0.0018 ($P< 0.001$) | 0.029 | 0.202 | 0.28 |
| SW | 0.0006 ($P= 0.191$) | 0.0007 ($P< 0.001$) | 0.0014 ($P< 0.001$) | 0.252 | 0.294 | 0.333 |
| TSW | 0.0001 ($P= 0.387$) | 0.0004 ($P= 0.009$) | 0.0028 ($P< 0.001$) | 0.128 | 0.513 | 0.125 |
| LL | 0.2164 ($P= 0.183$) | 0.1909 ($P\leq 0.001$) | 0.1881 ($P< 0.001$) | 5.556 | 6.783 | 0.504 |
| LW | 0.0510 ($P= 0.191$) | 0.0610 ($P\leq 0.001$) | 0.0598 ($P< 0.001$) | 3.966 | 4.743 | 0.505 |
| EO | 0.0004 ($P= 0.235$) | 0.0016 ($P< 0.001$) | 0.0019 ($P< 0.001$) | 0.128 | 0.513 | 0.457 |

RESULTS

Performance characterization of natural populations of myrtle

Descriptive analysis of morphological traits and essential oil content of three natural populations of Moroccan *Myrtus communis* L., are presented in (Table 3). The results indicated that the mean for the most morphological traits (e.g. FL= 0.87 cm, FWD= 0.72 cm, FWG= 0.23 g, TSW= 0.087 g, LL= 3.8 cm, LW= 1.6 cm) and oil essential content (EO~ 0.34 %), was highest in the Chefchaouen population from Rif Occidental, as compared to Taounat (from Rif Oriental) and Rabat (from Central Oceanic Atlantic) populations. With the exception for seed number (SN) and seed width (SW) which the population of Chefchaouen exhibited the medium values; SN= 6.64 and SW= 0.23 cm in Chefchaouen vs SN= 7.91 in Rabat population and SW= 0.27 cm in Taounat, respectively. Differences among populations were less clear for seed traits than for fruit and leaf traits. However,

these myrtle populations did not show highly significant differences in all studied traits ($P\geq 0.183$) (Table 4), as shown by MIXED procedure with the restricted maximum likelihood method.

Assessment of morphological and essential oil content variability

Estimates of variance components, coefficient of variation and broad-sense heritability for morphological traits and essential oil content are presented in (Table 4). Results of the analysis of variance of the various traits of natural populations, revealed that the population variance ($\hat{\sigma}_p^2$) was negligible to low. This indicated that the differences among populations for all measured traits were lowest. Relative to among populations variance, within population variance ($\hat{\sigma}_f^2$) contributed very significantly to the genetic variance of the different traits under investigation ($P\geq 0.05$), except for fruit weight ($P= 0.30$). This within

population variability was ranged from 0.20 % (seed length) to 79 % (number seed) of the total variance. Within population variability revealed remarkable features for different traits (except fruit weight) in Moroccan myrtle. Moreover, the results showed that the residual variance ($\hat{\sigma}_e^2$) was also an important part of the phenotypic variance.

Regarding to the broad-sense heritability (H^2), fruit length and width exhibited the highest values; 0.881 and 0.980, respectively. While the lowest H^2 value was related to the fruit weight (0.031), total seed weight (0.125), seed length (0.280), and seed width (0.333). The medium values' heritability were related to seed number (0.439), essential oil content (0.457), leaf length (0.504), and leaf width (0.505).

Correlation between morphological traits, essential content and geographical structure

Phenotypic correlations coefficient between morphological traits, essential oil and ecological conditions are presented in (Table 5).

Morphological traits correlations

Quantitative traits of myrtle's fruit revealed a high and significant phenotypic correlation; between fruit length and fruit width ($r= 0.695$) and fruit weight ($r= 0.771$). Moreover, a high and significant correlation was observed between fruit width and fruit weight ($r= 0.775$). Similarly, a significant and highly positive correlation was observed between leaf length and leaf width ($r= 0.808$). For quantitative seed traits, the correlations were less significant between seed traits than for fruit and leaf traits. However, seed number was negatively and moderately correlated with seed length and width ($r= -0.361$, $r= -0.373$; respectively). Positive and moderate correlations were also found between between seed length and width ($r=0.380$).

Both fruit width and weight were correlated with seed number ($r= 0.371$, $r= 0.366$; respectively), total seed weight ($r= 0.396$, $r= 0.437$; respectively), and marginally correlated with leaf length ($r= 0.286$, $r= 0.225$; respectively). There is also a weak correlation between fruit width and leaf width ($r= 0.240$). In addition, the fruit length exhibited moderate phenotypic correlations with seed width ($r= 0.313$) and total seed weight ($r= 0.312$).

Correlation between morphological traits and essential oil content

Essential oil content exhibited a moderate phenotype correlation with fruit width ($r= 0.361$). A marginal phenotypic correlation was also noted between essential

oil content and fruit length ($r= 0.219$). In the other hand, there was a negative marginal correlation between essential oil content and seed length ($r= -0.245$).

Geographical structure of morphological traits and essential oil content

At geographical structure of morphological traits and essential content levels, non-significant correlation was noted for most of these traits, except for leaves traits, seed width, seed number and essential oil. Leaves traits are related significantly to altitude, longitude north and to precipitation (ranged from 0.434 to 0.577). The latitude west was moderately correlated with seed number ($r= 0.316$), seed width ($r= -0.395$), and leaf length ($r= -0.318$). Also, moderate correlation was noted between essential oil content and precipitation ($r= 0.342$).

DISCUSSION

This study sheds light on the sources of phenotypic variance in morphological traits and essential oil content of Moroccan natural populations of *Myrtus communis* L. This study was also not designed to determine what biotic or abiotic environmental factors caused this variation, but was carried out to evaluate the effect of environmental on these traits, referred to geographical structure, and then speculate on what factors are most likely. Thus, measures of morphological traits and essential oil content were performed on a large number of individuals of three natural populations.

Performance of natural populations of myrtle for some parameters genetic

The results showed a low level of morphological traits and essential oil content variability among the three Moroccan natural populations of myrtle, but the significant variation resided within population of most traits. These suggested a uniform of genetic and genetic x environment interaction among myrtle populations. The lower variability of trait, measured in this study, among populations was different from our expectations. We expected that among populations variation would be the largest, based on most previous studies reported in this species (Mulas *et al.*, 2002; Ruffoni *et al.*, 2003; Messaouda *et al.*, 2007; Barboni *et al.*, 2010; Migliore *et al.*, 2012). These unexpected results let us to suggest that environmental conditions of locations of this species (Rabat, Taounat and Chefchaouen), which vegetal material was sampled, may not have an extreme and large variation and that phenotypic plasticity is likely controlled by random environmental micro-variations, both in space and time. As a consequence, no significant

Table 5. Correlation matrix among morphological traits, essential oil content and ecological conditions of three natural populations of Moroccan *Myrtus communis* L.. * Al is Altitude, LN is longitude north, LW is latitude west, and P is precipitation.

| Characters studied | FL | FWD | FWG | SN | SL | SW | TSW | LL | LW | EO |
|------------------------------|--------|-------|--------|--------|--------|--------|-------|--------|--------|-------|
| <i>Fruit traits</i> | | | | | | | | | | |
| FL | 1.000 | | | | | | | | | |
| FWD | 0.695 | 1.000 | | | | | | | | |
| FWG | 0.771 | 0.775 | 1.000 | | | | | | | |
| <i>Grain traits</i> | | | | | | | | | | |
| SN | 0.142 | 0.371 | 0.366 | 1.000 | | | | | | |
| SL | 0.205 | 0.135 | 0.117 | -0.361 | 1.000 | | | | | |
| SW | 0.313 | 0.075 | 0.193 | -0.373 | 0.380 | 1.000 | | | | |
| TSW | 0.312 | 0.396 | 0.437 | 0.296 | 0.003 | 0.025 | 1.000 | | | |
| <i>Leaf traits</i> | | | | | | | | | | |
| LL | 0.075 | 0.286 | 0.225 | 0.033 | 0.095 | 0.064 | 0.115 | 1.000 | | |
| LW | -0.073 | 0.240 | 0.104 | 0.080 | 0.050 | 0.016 | 0.075 | 0.808 | 1.000 | |
| <i>Essential oil content</i> | | | | | | | | | | |
| EO | 0.219 | 0.369 | 0.192 | 0.194 | -0.245 | -0.084 | 0.064 | 0.114 | 0.122 | 1.000 |
| <i>Ecological conditions</i> | | | | | | | | | | |
| Al | 0.157 | 0.256 | 0.250 | -0.207 | 0.178 | 0.109 | 0.057 | 0.577 | 0.412 | 0.156 |
| LN | 0.108 | 0.203 | 0.194 | -0.167 | 0.128 | 0.066 | 0.060 | 0.551 | 0.434 | 0.286 |
| LW | -0.048 | 0.084 | -0.073 | 0.316 | -0.165 | -0.359 | 0.050 | -0.318 | -0.101 | 0.074 |
| P | 0.112 | 0.250 | 0.204 | -0.118 | 0.109 | -0.007 | 0.078 | 0.556 | 0.472 | 0.342 |

differences among populations for traits measured in this study are likely to be due to micro-environmental variation effects, although genetic differences cannot be ruled out completely. A lot of effort is specifically put into finding an appropriate balance between inbreeding and out breeding depression (Avisé, 1992), but large phenotypic plasticity could compensate local adaptation or low genetic variability. No study has quantified the genetic diversity and structure of natural populations of Moroccan myrtle using neutral markers. The weak genetic differentiation may account for the lack of myrtle populations variation for morphological traits and essential oil content found in this study. For any cases, absence or weak genetic differentiation among populations is often explained by (i) low levels of genetic variability at the metapopulation (Giles and Goudet 1997; Noel *et al.*, 2007), (ii) high gene migration rates that homogenize populations and prevent differentiation. For example, previous studies has suggested the presence of long-distance gene flow in wild radish, perhaps through contamination of agricultural seed with wild radish seeds, which could reduce genetic differentiation over long distances (Kercher and Conner, 2003). (iii) or by homogenizing selection which the generalist suite of pollinators causes similar selection on floral traits in both locations, and then will have impact on cycle life of plant and then on fruits, seeds and leaves production (Jennifer and Jeffrey, 2001). In addition, number of individuals sampled per population and/or populations considered of this study, could also be responsible for a low genetic variability (Paland and Schmid, 2003; Noel *et al.*, 2007). Otherwise, small sample sizes would be made impossible to properly estimate differences among populations due

to biased weighting. Nevertheless, more individuals and more populations should be considered for future study to confirm this result.

A significant high variation within population for all morphological traits (except fruit weight there isn't variation within and among populations) and essential content. These results confirmed the differences in the genotypes genetics potential, and indicated that certain genotypes carried alleles with different additive effects. This observed within-population variability in morphological traits and essential content may give advantage to the populations of *Myrtus communis* L. under the unpredictable environmental conditions. The difference between genotypes (within population) in morphological traits and essential oil content could be influenced by genotype, environment and interaction among them (Lopez *et al.*, 2008). A possible explanation for why there was more variation within population than among populations is due to difference in fruits, seeds and leaves age (Lopez *et al.*, 2008). Otherwise, fruits, seeds and leaves of two adjacent plants on the same location are almost always different in age. Wannes *et al.* (2007) stated that essential oil yield varied in leaves, fruits and stems of *Myrtus communis* varieties (italica and baetica). So, in leaves, it was 0.5% for italica and 0.3% for baetica and was higher than in fruits and stems with respectively, 0.1% and 0.04% for italica and 0.07% and 0.03% for baetica. The within population variation could be also caused by changes in inter- or intra- plant competition in the same location over the season (Jennifer and Jeffrey, 2001). Another possible biotic environmental factor is herbivory, since Strauss *et al.* (1996) found that herbivory affects variation among plants

in petal size. Moreover, animal pollinators cause natural selection on floral traits in many species (Cambell, 1989). This pollinator-mediated selection acts on phenotypic variation among individuals in a population (Jennifer and Jeffrey, 2001).

Also, respond plastically to environmental variations and phenotypic plasticity should be considered a major evolutionary force, especially in plants, which cannot escape their local environment (Ohsawa and Ide, 2008). So the vast majority of the phenotypic variation within population in morphological traits and essential oil content would be due to the environment and plant ontogeny.

The characterization of geographical pattern of morphological variation and essential oil content in natural populations suggests possible patterns of polymorphism genetic and plastic responses to environmental gradients (Wannes *et al.*, 2007). This is because individual plant develops the ability to produce different phenotypes as a response to abiotic stress, often referred to phenotypic plasticity (Wahid *et al.*, 2012).

These plasticity responses are expressed at different levels such as plant morphology, anatomy, physiology and chemical secretion (Wannes *et al.*, 2007; Wahid *et al.*, 2012). A further motivation for understanding sources of morphological traits and essential oil content variation within and among populations can help to explain differences between environments in heritabilities of these traits.

The present study showed variation of broad sense heritability estimates among traits. These differences could be due to the difference in the expression of genes that control these traits (Wahid *et al.*, 2012). The characters namely; fruit length and width ($H^2= 0.88$ and 0.98 , respectively) displayed the highest heritability, and seed number ($H^2= 0.44$), seed width ($H^2= 0.33$), leaf length ($H^2= 0.50$), leaf width ($H^2= 0.51$) and essential oil content ($H^2= 0.46$) displayed the medium values of heritability estimates.

But the lowest heritabilities were reserved to fruit weight, seed length and total seed weight (ranged between 0.03 and 0.12). The results showed that genetic factors were more effective than environment on components. In fact, high and medium heritabilities indicate that these traits are less affected by environmental heterogeneity than other traits, and the control of additive gene and selection may be effective for those traits (Rajput *et al.*, 2004; Wahid *et al.*, 2012; Abou El-Nasr *et al.*, 2013). Although the broad sense heritabilities appear high or medium for most traits, it is well known that these heritabilities are populations and sites dependent and that consequently they could change depending on the group of populations other than those considered in this study, the age of the vegetative material, and the environment in which the genotype will be tested.

Correlation and geographic structure of morphological traits and essential oil content

A strong and positive correlation was found, in this study, between most morphological traits (fruit, leaf and some seed traits) and between essential oil content. Among traits, fruit width appears to be correlated to length fruit and weight, seed number and total weight seeds, and leaf length and width. These results lead to the conclusion that an individual selection for fruit width, either positive or negative, may produce similar results for length of fruit and weight, seed number and total weight of seeds, and leaf length and width. A recommend selection of superior genotypes in natural population based on fruit width is more suitable because in addition to the strong positive genotypic correlation between the most morphological traits, fruit width is under stronger genetic control. Moreover, a moderate correlation between fruit width and essential oil indicates that biggest fruit contribute for sufficient volatile compounds in myrtle's leaves. Therefore, the fruit trait, essential oil content and their associated traits are an important character for Moroccan myrtle and can be good criteria for selection at the segregating generations in the future plant breeding programs.

Environmental conditions appear to be important determinants of the morphological traits (Wahid *et al.*, 2012). Leaves morphology traits appear to be linked to variations in water availability (precipitation) and temperature stress tolerance related to altitude and longitude north. Our results suggest that the population of *Myrtus communis* L. in the higher Occidental Rif (Chafchaouen) might have adapted to humid conditions as it is characterized by longer leaves compared to similar population (Taounat) at Oriental Rif, characterized by lower longitude north with high temperature and moderate humidity regimes. Also, moderate correlation was noted between essential oil content and precipitation, which the most humid zones (Occidental Rif and Central Oceanic Atlantic) is characterized by the high essential oil content.

Correlation analysis between the chromosomes number and production ability of the natural populations is a new line of investigation for medicinal and aromatic plants. In this respect, studies concerning *Marrubium vulgare* (Letchamo and Mukhopadyay, 1997), *Acorus calamus* (Rode *et al.*, 1996) and 31 other species by Murin (1997) are worth to mention.

Conclusion

The significant variation resided among individuals of most traits studied, indicating that individual plants responded differently to the variable environment and parental effects. However, most of different traits measured of individual myrtle plants exhibit a high or

medium broad sense heritability estimates, confirming that variation among individuals are relating more to additive genetic. Comparing with all traits, fruit and leaf traits and essential oil content presented more heterogeneity and high to medium heritability. Therefore, these traits are an important indicator for Moroccan myrtle performance and be good criteria for selection at the segregating generations in the future plant breeding programs. A clinal gradient (altitude, precipitation and longitude) of morphological variation leaf traits and essential oil content is showed and suggested an important role of natural selection and adaptation to a rapidly changing environment in the establishment of these patterns. Moreover, strong and positive correlation was found, in this study, between most morphological traits (fruit, leaf and some seed traits) and between essential oil content. Comparing all traits, fruit trait, essential oil content and their associated traits are an important character for Moroccan myrtle and can be good criteria for selection at the segregating generations in the future plant breeding programs.

AUTHORS` DECLARATION

We declare that this study is an original research by our research team and we agree to publish it in the Journal.

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