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## To Study of Essential Oil and Agricultural Properties of Vetiver (*Vetiveria Zizanioides*) in the Southeastern of Mediterranean

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### ABSTRACT

Vetiver (*Vetiveria zizanioides*), a plant of the *Gramiceae* family, is well known as an eco-friendly tool to prevent soil erosion, and one of the most important raw materials in perfumery industry. Its roots are usually steam distilled to obtain vetiver oil. The complex odor profile of vetiver oil is dominated by a woody and special balsamic tonality. This study was conducted to investigate determination of essential oil content and composition and agricultural properties of Vetiver grown at the experiment area of the Field Crops Department, Çukurova University, Agriculture Faculty, during 2008 and 2009 under Çukurova ecological conditions. Plant height, number of tiller, fresh and dry herbage yield, fresh and dry root yield, essential oil content of dry root, and composition of essential oil by GC-MS were determined during autumn. Twenty eight components were detected in the essential oil from cultivated vetiver. Khusinol (19.15% and 15.67%), b-vetivenene (9.76% and 8.16%) and dehydro-aromadendrene (7.34% and 9.66%) in the experiment years were present as the major components in the essential oil.

**Kew words:** vetiver, root yield, essential oil, components, khusinol.

### Introduction

Vetiver (*Vetiveria zizanioides*), a plant of the *Gramiceae* family, is well known as an eco-friendly tool to prevent soil erosion, and one of the most important raw materials in perfumery industry. Its roots are usually steam distilled to obtain vetiver oil. The complex odor profile of vetiver oil is dominated by a woody and special balsamic tonality. This oil is much appreciated by the perfume industry, where it is used as a fixative and as a odor contributor in bases, such as rose, chypre, and in several masculine fragrances. Furthermore, vetiver can be used in the prevention of soil erosion by rain and in the

combat against soil contamination by heavy metals. Its oil can be applied in aromatherapy and in food, as aroma in canned asparagus and peas and as flavor agent in some beverages [1].

Vetiver grass can be used at the steep slopes of arid regions where erosion is severe to prevent erosion due to the fact that it has proven successful in holding the soil [2]. Because it has short rhizomes and a massive, finely structured root system that grows very quickly; this deep root system makes the vetiver plant extremely drought tolerant and dislodge when exposed to a strong water flow [3].

The vetiver essential oil production is closely related to the metabolism of plant roots, which

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is affected by changes in environmental temperatures such as temperature, during winter the drop in temperature causes a decrease in metabolic plant activities, which in turn slows down oil production [4]. Among the odorous components in vetiver oils from different sources, khusimol, b-vetivone, a-vetivone are the major constituents, and their presence is often considered as the fingerprint of the oil [5].

Although vetiver is widely cultivated in tropical and sub-tropical regions, is not commonly grown in Turkey. Limited studies have been conducted to determine erosion control of sloppy areas [2]. Likewise, the essential oil content and composition of vetiver are unknown our conditions. For this reason, this study was conducted to investigate determination of essential oil content and composition and agricultural properties of vetiver conditions in the Southeastern of Mediterranean conditions.

### Material and Methods

This study was conducted under Çukurova ecological conditions (latitude 37°01'05 N and longitude 35°21'33E, 73 m above sea level) at the department of Field Crops, Faculty of Agriculture, Cukurova University (Adana, East Mediterranean Region of Turkey), during 2007-2008 and 2008-2009 growing seasons.

Vetiver (*Vetiveria zizanioides* Nash.) plants from Karadeniz Technical University, Faculty of Forestry were transplanted on seedbed in the greenhouse in the November, 6 in 2006 at experiment area of Department of Field Crops. The plants left for sprouts over one year. Plants were cut 25-30 cm above ground and dug out for preparation of propagation materials. Plants were divided into slips with 1-2 tillers. These tillers were transplanted in the open field plots in rows 50x50 cm in 26, February 2008. Field trials were arranged in completely randomized block design with 3 replications, each plot consist of four plants. The plots were given 40 kg ha<sup>-1</sup> N and 20 kg ha<sup>-1</sup> P<sub>2</sub>O<sub>5</sub> before planting. During planting, the plants were irrigated immediately, later, they were irrigated when needed during two years. In the second year, the plant was cut of for its aerial parts before winter. The plants were dug out in September in both years. Plant height and number of tillers in each plot were determined at the harvest dates. Vetiver height was determined as the length of the longest leaf in each plant. Fresh and dry herbage yields, fresh and dry root yields, essential oil content of dry root were measurement. The herbage and root were dried under shade at an airy place for one week.

The soil of experimental area is classified as a sandy loam in the upper 0-30 cm profile, which contained an average 1.3% organic matter with pH 7.11, 478 K mg kg<sup>-1</sup>, 15 P mg kg<sup>-1</sup>, 0.69 Zn mg kg<sup>-1</sup>, 12.4 Mn mg kg<sup>-1</sup>, 1.26 Cu mg kg<sup>-1</sup> and 9.6 Fe mg kg<sup>-1</sup>.

Mediterranean climate prevails in this region. The warmest months are July and August, the coldest month is January. Nearly 90 % of annual rainfall falls in the winter months from December to April. Evaporation losses and relative humidity are high in summer. The data taken from the State Meteorology Institute, Adana (Turkey) showed that climatic conditions varied slightly between the experimental years. Mean temperatures were 19 °C in both years. The highest temperatures of first and second years were 42. °C in the August and 38.4 °C in the June respectively. Total precipitation for 2008 and 2009 until cutting dates were 156.2 and 294.1 mm, respectively, rainy days were usually between December and March. January was the coldest month (-1.1 and -2.0°C ) in both years.

### Extraction:

The essential oil of dried root (30 g) was extracted by hydro-distillation for 3 h under continuous steam using a Clevenger apparatus [4]. The essential oils stored in glass vials and kept at ± 4 °C until chemical analysis.

### GC-MS Analysis:

Qualification and quantification were carried out by using a Finnigan-Trace GC-MS equipped with an auto sampler. One microlitre of sample volume was injected using split method with 50 split ratio. Chromatographic separations were accomplished with a Zebtron ZB-5 capillary column (5% phenyl-95% dimethylpolysiloxane, 0.25mm i.d.×60 m, film thickness 0.25 μm). Analysis was carried out using helium as the carrier gas, flow rate 1.0 ml per minute. The column temperature was programmed from 50 to 240 °C at 3 °C min<sup>-1</sup>. The injection port temperature was 250 °C. The ionization voltage applied was 70 eV, mass range *m/z* 41-400 a.m.u. The separated components were identified tentatively by matching with GC-MS results of National Institute of Standards and Technology (NIST) 05 mass spectral library data because their reference reagents were not available. The quantitative determination was carried out based on the peak area integration.

**Statistics:**

The data were statistically analysed by computing MSTAT-C package program with randomized block design.

**Results and discussion**

Vetiver was grown in Adana, East Mediterranean Region of Turkey, there is no previous existing work about it in the region. The results of two years for vetiver observed are given in Table 1. Plant height was not affected years significantly. It was varied between 144.43 and 146.00 cm in 2008 and 2009, respectively.

According to Dudai *et al.* [3], vetiver plants can survive in summer in semiarid regions around the Mediterranean under rain-fed condition, but their height will be reduced, they reported that the heights of the irrigated and rain-fed plants were 2.5 and 2.0 m, respectively but, it was shorter after foliage-cut treatments were 1.5-1.9 m. The plant heights in our study are shorter and some way similar than the finding of researches mentioned above.

Vetiver can be propagated by tiller, it was a typical growth pattern of plants. Numbers of tiller per plant, at two years of vetiver plants are presented in Table 1. No significant differences were found among years in numbers of tiller per plant, however, second year's (56.10) result was higher than the first year (43.67). Edelstein *et al.* [6] said that the number of sprouts per plants were varied between 23 and 41.2 with three fertilizer pot treatments. It reached till over 40 per plant with increasing minimum-maximum air temperatures, 21 and 29° C, respectively [3]. Our results are similar, and values are higher than the finding of researches mentioned above.

Although vetiver has massive, complex and long (3-4 m) root system [7], in our study, root length of first and second year varied 22.5 and 30.3 cm, respectively. However, the economic part of vetiver is its dug up to a depth of 30-40 cm, yields and oil of better quality obtained from thicker roots with 15-18 months crop [8]. Therefore, our results are suitable for this the best and economic crop.

Years did not affect fresh herbage and dry yield significantly (Table 1). The highest fresh and dry herbage yield were recorded in the second year. Yield values of 2009 were higher than that of 2007. Vetiver is a high biomass plant having high C4 photosynthetic efficiency [7], for this reasons, it can be produced biomass mean 51 t ha<sup>-1</sup> fresh herbage yield and 29 t ha<sup>-1</sup> dry herbage yield. Fresh root yield was not affected by years significantly. Mean of years

was 3.8 t ha<sup>-1</sup>. Years affected dry root yield significantly (Table 1). The highest dry root yield was recorded in the second year. Yield values of 2009 was higher than that of 2008, because plant is perennial. The age, the soil, climatic conditions and the strain are important factors governing the yield of roots. Average of 3-4.5 t ha<sup>-1</sup> dry root was obtained under good management [8]. In India, root yield of vetiver varied between 1.27 and 2.52 t ha<sup>-1</sup> according to the location and Bettaland types [9]. These values are in agreement with our second year result.

The essential oil content of vetiver obtained from years were given in Table 2. Essential oil content of vetiver was changed between 0.78% (v/w) and 0.70% (v/w) in the first year and the second year. Literature review showed big variation between the essential oil content of vetiver. For example, the essential oil content of vetiver were between 0.06% and 0.27% in different cultivation systems [10], 0.36% and 1.38% in three operating parameters such as pressure, temperature and time, of supercritical carbon dioxide extraction methods [11], 0.6 % and 3.2% in methods and times of extraction [1] or 0.38% and 0.7 % in cold and warm seasons [4]. However, some researchers reported that essential oil content under open condition is significantly lower then compared to that in shade, Shade grown plants showed higher volatile oil content [9]. From these results, we can conclude that both environmental and extraction methods are effective in the production of essential oil.

Essential oil of vetiver is composed of more than 100 components that are mainly sesquiterpenes and their derivatives. The main constituents of vetiver oil comprise of: sesquiterpene hydrocarbons and their alcohol derivatives- vetiverols such as, khusimol, khusinol, carbonyl derivatives-vetivones (ketones) such as, vetivone, khusimone and three carbonyl compounds, such as  $\beta$ -vetivone,  $\alpha$ -vetivone and khusimone [12]. Among the odorous components in vetiver oils from different sources, khusimol,  $\beta$ -vetivone,  $\alpha$ -vetivone are the major constituents, and their presence is often considered as the fingerprint of the oil, are also present in the oil giving characteristic odour of Vetiver oil [5]. Also, economically important active principles of vetiver were  $\alpha$ -vetivone (2.19-5.53%),  $\beta$ -vetivone (3.12-4.82),  $\beta$ -eudesmol (6.44-8.06) and khusimol (14.92-26.94) [9].

The chemical components of the essential oil obtained from dry root of vetiver on different years was indicated in Table 2. Twenty eight components were identified in the both experiment years, representing from 79% in the essential oil.

Khusinol,  $\beta$ -vetivenene and dehydro-aromadendrene were main components in both years. In the experimental years; the highest values of khusinol obtained from the first year, and it was varied between 19.15% and 15.67% in the first year and second year. Adams *et al.* [13], reported that khusinol is the largest component of vetiver oil among thirteen accession between locations (Nepal, Portugal and Florida), its contents varied

from 14.52% to 31.42%. In addition, it was indicated that the percent composition of khusinol is less variable.  $\beta$ -vetivenene contents varied 9.76% and 8.16% first and second years, respectively. Carbonyl compounds,  $\beta$ -vetivone and  $\alpha$ -vetivone contents in first and second year were 2.64% and 2.55% and 3.17% and 3.54%, respectively. Our results are similar to the finding of researches mentioned above.

**Table 1:** Effect of years on plant height (cm) and fresh and dry herbage yields (t ha<sup>-1</sup>) of vetiver.

Years	Tiller	Root	Lengh (cm)	Yields (t ha <sup>-1</sup> )			
	Plant height (cm)	numberPer plant		Fresh Herbage	Dry Herbage	Fresh Root	Dry* Root
2008	144.43	43.67	22.53	46.67	28.00	1.73	0.83
2009	146.00	56.10	30.30	56.80	30.23	5.87	3.30
Mean	145.22	49.89	26.42	51.74	29.12	3.8	2.07
LSD (5%)	ns	ns	ns	ns	ns	ns	2.728
CV (%)	8.41	19.15	15.69	22.73	38.37	46.53	30.67

\*Means followed by the same letter are not significantly different (P< 0.05), ns: non significant

**Table 2:** Essential oil content and its chemical constituents of vetiver during 2008 and 2009.

Parameters	Years	
	2008	2009
Essential oil content (%)	0.78	0.70
RT	components	
55.85	8,9-dehydro-cycloisolongifolene	1.83
56.54	cadina-1(10),4-diene	1.12
56.91	ar-curcumene	2.72
57.27	$\gamma$ -cadinene	0.49
59.99	$\beta$ -vetivenene	9.76
60.54	$\gamma$ -gurjunene	1.34
61.82	ledene oxide	0.47
62.56	$\eta$ -himachalene	0.14
63.02	valencene	0.18
63.51	C15H22O	0.64
63.99	$\gamma$ -muurolene	2.85
64.63	b-eudesmol	0.33
64.78	$\beta$ -guaiene	0.43
65.09	allaromadendrene oxide	0.96
65.44	$\delta$ -cadinol	2.59
65.77	junipene	0.44
66.01	tau-muurolol	1.99
66.24	trans-caryophyllene	3.69
66.50	cubenol	0.59
66.86	caryophyllene oxide	1.66
67.27	khusinol	19.15
67.45	8-cedren-13-ol	2.72
68.68	C13H18O	3.18
70.06	$\beta$ -gurjunene	3.20
71.97	thujopsene-13	3.80
73.47	dehydro-aromadendrene	7.34
75.01	$\beta$ -vetivone	2.64
76.52	$\alpha$ -vetivone	3.17
	Total identified	79.42
		79.93

**Conclusion:**

As a result, vetiver was growing first time in the Southeastern Mediterranean province. Essential oil content and its chemical constituent have been reported the first time. Results of root yield, content of essential oil and components of essential oil were very hopeful for its future production and adaptation to the location.

Moreover, we have to find good cultivation conditions and suitable processing for essential oil distillation for high root yield and good oil quality and high oil productivity.

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