# Influence of Vermicompost and Harvest Time on some Characteristics of Hyssop (*Hyssopus officinalis* L.)

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## ABSTRACT

Hyssop (*Hyssopus officinalis* L.) is herbaceous perennial plants of Lamiaceae family grown in Europe, the Middle East, Asia, and Northern Africa. Field experiment was carried out during 2015/2016. The experimental design was laid out as factorial in randomized complete block design that five levels of fertilizer treatment (0, 5, 10, 15 ton ha$^{-1}$ vermicompost and N 75 kg ha$^{-1}$ source of urea and three harvesting time (pre-flowering, blooming and full flowering stages) were arranged in experimental plots. Results indicated that percentage of essential oil varied from 0.474 to 0.585 in different stage of plant growth. Vermicompost levels had a promoting influence on most of growth traits and accelerated essential oil accumulation. With enhancing levels of vermicompost essential oil content increased and use of vermicompost in high level (15 ton ha$^{-1}$) produced the maximum (18.46 kg ha$^{-1}$) essential oil yield. Similarly, the forth level of vermicompost (15 ton ha$^{-1}$) had significantly effect on Chlorophyll, carotenoid, total flavonoids and anthocyanins. The greatest plant high, stem diameter, number of secondary and flowering branches as well as dry weight was gained for plant grown in high levels of vermicompost. Overall, we conclude that utilization of 15 ton ha$^{-1}$ vermicompost and harvest Hyssop at full flowering stage is recommended for optimizing the content of essential oils and photosynthetic pigments and morphological characteristics in Hyssop.

**Keywords**: Chlorophyll, essential oil, *Hyssopus officinalis*, vermicompost

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## INTRODUCTION

Hyssop (*Hyssopus officinalis L.*), which is known as “Zufa” in Persian, is herbaceous perennial plants belong to Lamiaceae family that widely grown essential oil crop in Europe, the Middle East, Asia, and Northern Africa (Tonutti and Liddle, 2010). The essential oil of hyssop is mainly used in perfumery, cosmetics, beverages, foods, and flavoring industries (Tonutti and Liddle, 2010) and extraction from the aerial parts are used for expectorant, carminative, anti-inflammatory, anticatarrhal and antispasmodic in traditional medicine in many parts of the world (Kizil *et al*., 2008). Extracts of the plant are also used for their antimicrobial, antioxidant properties (Kreis *et al*., 1990; Letessier *et al*., 2001) and exhibit strong antiviral activity against HIV (Ozer, *et al*., 2006). According to Kohlmunzer (2007) in hyssop percentage of essential oil was 1% while Zawislak (2011) reported that essential oil of Hyssop ranged from 0.67 to 1.65% in vegetative and full blooming stages respectively. The essential oil biosynthesis is mainly attributed to genetic factors, but other factors have important effect on production of essential oil such as climate and soil condition, harvesting time, development stage and plant age (Moraes, 2009). In this regard Nemeth *et al*. (2000) demonstrated that the essential oil contents in Hyssop significantly changes during plant growing. Ozguven and Tansi (1998) reported that the greatest essence content of *Thymus vulgaris* plants obtained at full flowering and post-flowering stages. Although the excessive utilization of chemical fertilizer enhanced crop productivity but it has irreversible effect on soil health and water resources such as pollution of surface and ground water resources (Townsend *et al*., 2008).

Therefore, organic fertilizers with decrement negative effects on human health and environment can be considered as an alternative to sustainable agriculture development. Vermicompost use not only has beneficial effects such as water maintenance capacity of soil, supply of nutrients, increment of organic matter, increase biological activity of soil and reduction of soil erosion but also encourage plant growth and development with better uptake nutrient elements by plant roots (Singh *et al*., 2010; Zhang *et al*., 2015). Numerous studies have been reported that vermicompost had favorable influence on quantity and quality of characteristic in medicinal plants such as Fennel (Darzi *et al*., 2009), coriander (Singh *et al*., 2009), basil (Anwar *et al*., 2005) and peppermint (Ayyobi *et al*., 2014). However, to the best of our knowledge, there is no sufficient information about impact of vermicompost and harvesting time in Hyssop. Therefore, the objective of the study was to determine the effects of different vermicompost levels and harvest time on essential oil, photosynthetic pigment and some morphological characteristics of Hyssop.

## MATERIALS AND METHODS

Field experiment was carried out during 2015/2016 at the Research Field of Payam Noor University of Marand, Iran (38°22' N, 45°46' E and 1500 m Altitude). The experimental design was carried out as factorial in randomized complete block design with three replicates. Five levels of fertilizer treatment (0, 5, 10, 15 ton $ha^{-1}$and chemical fertilizer or N 75 kg $ha^{-1}$ source of urea) and three harvesting time (pre-flowering, blooming and full flowering stages) were arranged in experimental plots. The composite soil samples were collected one week before planting and some of physicochemical properties were evaluated. Details of the properties of soil and vermicompost are shown in Tables 1 and 2. The total vermicompost and half of the total N applied were scatted by hand and mixed into the soil five days before planting. With attention soil analysis K and P fertilizers were not used. The remaining half of the N was applied as top dressing when the Hyssop seedlings were at the eight-leaf stage. Each plots had six rows with 3 m long that was spaced 0.50 m apart. To eliminate any influence of lateral water movement 1.5-m alley was maintained between all plots.

Seedlings of Hyssop were planted by hand on 10 May 2015. Weeds were controlled by hand three times after the planting. The plants were harvested in pre-flowering, beginning flowering and full flowering stages, on 15 June, 27 July and 10 August 2015 respectively. At each stage data were recorded for plant and studied traits included: plant height (PH), stem diameter (SD), Number of secondary branches (NSB), number of flowering branches (NFB), essential oil content (EOC), dry weight (DW), essential oil yield (EOY), total anthocyanins (TA), chlorophyll a (CA), chlorophyll b (CB), total chlorophyll (TC), carotenoid (C) and total flavonoids (TF). Photosynthetic pigments included chlorophyll (a, b and total) determined by (Arnon,1967). Total anthocyanins and total flavonoids measure by Krizek *et al*. (1993).

Analysis of variance and mean comparisons were performed using the GLM procedure of SAS (SAS, 2002). The least significant difference (LSD) method at a probability level of 0.05 was used to determine significant differences among means. Single-degree of freedom orthogonal contrasts were performed to separate differences vermicompost (F1 to F4) vs. chemical fertilizer (F5). For number of flowering branches, the square root transformation was performed.

**Table 1** Soil physical and chemical properties

| Soil texture | Ec ($dSm^{-1}$) | pH | Organic carbon (%) | N (%) | P ($mg\ kg^{-1}$) | K ($mg\ kg^{-1}$) |
|---|---|---|---|---|---|---|
| Sandy loam | 1.09 | 7.66 | 1.2 | 0.06 | 47 | 605 |

**Table 2** Chemical characteristics of vermicompost

| Soil texture | Ec ($dSm^{-1}$) | pH | Organic carbon (%) | N (%) | P (%) | K (%) | Fe ($mg\ kg^{-1}$) | Zn ($mg\ kg^{-1}$) |
|---|---|---|---|---|---|---|---|---|
| Sandy loam | 1.09 | 5.66 | 18.9 | 1.61 | 0.6 | 1.45 | 984 | 124 |

## RESULTS AND DISCUSSION

According to analysis of variance (Table 3), fertilizer was significant for plant height, dry weight, essential oil content, and essential oil yield at 0.01% probability while it was significant for stem diameter and number of secondary branches at 0.05% probability level. The harvesting time had significant effect on plant height, stem diameter, number of secondary branches, number of flowering branches, dry weight, essential oil yield (at 0.01% probability level) and essential oil content (at 0.05% probability level). The fertilizer × harvesting time interaction was significant only for plant height at 0.05% probability level of Table 3 and Figure 1. The results of the ANOVA for the other remained traits (Table 4), showed that fertilizer was significant for chlorophyll a, total chlorophyll and total Flavonoids at 0.01% probability level and for chlorophyll b, total carotenoid and total anthocyanins at 0.05% probability level. Also, harvesting time was significant for chlorophyll b and total chlorophyll at 0.01% probability level and it was significant for total anthocyanins, and total Flavonoids at 0.05% probability level (Table 4). The interaction between fertilizer and harvesting time was significant only for total anthocyanins at 0.05% probability level (Table 4). Whenever a two-way interaction was not significant for a given trait, the main effects are presented and explained. F-values for orthogonal comparisons between vermicompost treatments (F1-F4) and urea (F5) were significant on plant height, stem diameter, dry weight, essential oil yield, carotenoid and total flavonoids (Table 5 and 6).

**Table 3** Analysis of variance for the measured morphological traits, essential oil content and essential oil yield of hyssop (*Hyssopus officinalis* L.)

| SOV | df | PH | SD | NSB | NFB | DW | EOC | EOY |
|---|---|---|---|---|---|---|---|---|
| Block | 2 | 0.5803 | 2.978 | 19.81* | 0.185 | 182901.07 | 0.00004 | 6.361 |
| Fertilizer (F) | 4 | 122.26** | 18.51* | 11.83* | 0.112 | 2261375.6** | 0.0182** | 138.43** |
| Harvesting (H) | 2 | 953.65** | 35.59** | 25.15** | 18.88** | 8084973.3** | 0.0078* | 299.18** |
| F*H | 8 | 32.99* | 8.87 | 3.32 | 0.2003 | 152416.1 | 0.00073 | 11.02 |
| Error | 28 | 13.51 | 5.28 | 4.500 | 0.086 | 171053.5 | 0.0020 | 6.13 |
| Vermicompost vs. urea | 1 | 352.35** | 20.51* | 10.31 | 0.002 | 1279421.1* | 0.003 | 66.26** |
| CV | | 9.13 | 22.84 | 19.72 | 18.09 | 16.74 | 8.56 | 18.33 |

SOV = Source of variation; DF = Degrees of freedom; CV = Coefficient of variation
**, * and ns are significant at 1 and 5% probability level and non-significant, respectively
Traits are: plant height (PH), stem diameter (SD), Number of secondary branches (NSB), number of flowering branches (NFB), essential oil content (EOC), dry weight (DW), essential oil yield (EOY) and essential oil yield (EOY)

**Table 4** Analysis of variance for the photosynthesis pigments of hyssop (*Hyssopus officinalis* L.)

| SOV | df | CA | CB | TC | C | TA | TF |
|---|---|---|---|---|---|---|---|
| Block | 2 | 0.00012 | 0.0035 | 0.0024 | 0.0013 | 0.376 | 1522.92 |
| Fertilizer (F) | 4 | 0.1403 | 0.0301* | 0.2921** | 0.0048* | 2.906* | 9874.58** |
| Harvesting (H) | 2 | 0.1046* | 0.0548** | 0.2932** | 0.00026ns | 2.852* | 8851.08* |
| F*H | 8 | 0.0047 | 0.0176 | 0.198 | 0.00048 | 1.904* | 656.99 |
| Error | 28 | 0.0240 | 0.0099 | 0.0426 | 0.0014 | 0.721 | 1920.54 |
| Vermicompost vs. urea | 1 | 0.07 | 0.003 | 0.10 | 0.008** | 1.85 | 33190.21** |
| CV | | 17.72 | 18.18 | 14.49 | 20.91 | 13.36 | 12.05 |

SOV = Source of variation; DF = Degrees of freedom; CV = Coefficient of variation
**, * and ns are significant at 1 and 5% probability level and non- significant, respectively
Traits are: chlorophyll a (CA), chlorophyll b (CB), total chlorophyll (TC), carotenoid (C), total anthocyanins (TA) and total flavonoids (TF)

**Table 5** Mean comparison of the morphological traits, essential oil content and essential oil yield of hyssop (*Hyssopus officinalis* L.) for vermicompost and chemical fertilizer

| Fertilizer | SD (mm) | NSB | NFB | DW (Kg ha$^{-1}$) | EOC (%) | EOY (Kg ha$^{-1}$) |
|---|---|---|---|---|---|---|
| F1 | 8.55$^{b}$ | 9.36$^{c}$ | 2.1$^{b}$ | 1857.9$^{d}$ | 0.474$^{c}$ | 8.81$^{c}$ |
| F2 | 10.65$^{ab}$ | 9.65$^{bc}$ | 3.25$^{ab}$ | 2133.3$^{dc}$ | 0.518$^{bc}$ | 11.08$^{bc}$ |
| F3 | 10.43$^{ab}$ | 11.43$^{ab}$ | 3.51$^{ab}$ | 2832.9$^{ab}$ | 0.573$^{a}$ | 16.43$^{a}$ |
| F4 | 11.97$^{a}$ | 11.61$^{ab}$ | 3.52$^{a}$ | 3090$^{a}$ | 0.585$^{a}$ | 18.46$^{a}$ |
| F5 | 8.71$^{b}$ | 11.71$^{a}$ | 2.88a$^{b}$ | 2438.73$^{bc}$ | 0.522$^{b}$ | 12.74$^{b}$ |

Traits are: plant height (PH), stem diameter (SD), Number of secondary branches (NSB), number of flowering branches (NFB), dry weight (DW), essential oil content (EOC) and essential oil yield (EOY). Fertilizer: F1: control, F2: 5 ton ha$^{-1}$, F3: 10 ton ha$^{-1}$, F4: 15 ton ha$^{-1}$ and F5: urea or N 75 kg ha$^{-1}$. The same letters in each column indicate that there is no significant difference between the means

**Table 6** Mean comparison of the measured morphological traits and essential oil content of hyssop (*Hyssopus officinalis* L.) for three levels of harvesting time

| Harvesting | SD (mm) | NSB | NFB | DW (Kg ha$^{-1}$) | EOC (%) | EOY (Kg ha$^{-1}$) |
|---|---|---|---|---|---|---|
| H1 | 8.97$^{b}$ | 9.40$^{b}$ | 0.00$^{c}$ | 1775.3$^{c}$ | 0.508$^{b}$ | 9.18$^{c}$ |
| H2 | 9.39$^{b}$ | 10.84$^{ab}$ | 1.21$^{b}$ | 2398.1$^{b}$ | 0.543$^{a}$ | 13.23$^{b}$ |
| H3 | 11.82$^{a}$ | 12.00$^{a}$ | 7.91$^{a}$ | 3238.3$^{a}$ | 0.551$^{a}$ | 18.10$^{a}$ |

Traits are: plant height (PH), stem diameter (SD), Number of secondary branches (NSB), number of flowering branches (NFB), dry weight (DW), essential oil content (EOC) and essential oil yield (EOY). Harvest time: H1: pre-flowering, H2: blooming and H3: full flowering. The same letters in each column indicate that there is no significant difference between the means

**Morphological Characteristics**

For all levels of vermicompost, full flowering stage was the best in plant height while pre-flowering stage produced the lowest plant height. Zawislak (2011) reported that the maximum plant height of Hyssop was gained on average 48.8 and 47.9 cm, at full flowering and full blooming phases respectively compare to vegetative and beginning of flowering stages. According by Roslon *et al.* (2002) height of plants for *Hyssopus officinalis* L. at the full blooming stage achieved the 70 cm. In fact, with passage of time and using of vermicompost and chemical fertilizer improved height of plant. Similar results stated that Ayyobi *et al.* (2014) in peppermint (*Mentha piperita* L.), Darzi *et al*. (2012) in anise (*Pimpinella anisum*) and Hadi *et al*. (2011) in *Matricaria chamomilla*.

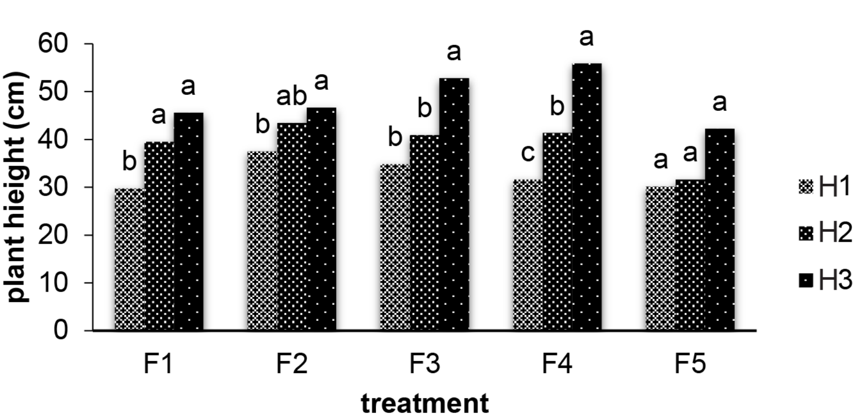


**Figure 1** The interaction between fertilizer and harvesting time on plant height of Hyssop Fertilizer: F1: control, F2: 5 ton ha$^{-1}$, F3: 10 ton ha$^{-1}$, F4: 15 ton ha$^{-1}$ and F5: urea or N 75 kg ha$^{-1}$. The same letters in each column indicate that there is no significant difference between the means. Harvest time: H1: pre-flowering, H2: blooming and H3: full flowering. The same letters in each column indicate that there is no significant difference between the means

The maximum stem diameter, number of secondary branches, number of flowering branches and dry weight was obtained using the F4 treatment, whereas the minimum value of these traits was obtained in control treatment. In addition, F4 treatment compare to chemical treatment (F5) significantly was better in mention characteristics. This finding may be attributed to better supply of macro and micro elements by vermicompost manure. Application of vermicompost with increase of organic matter, enzymatic activities, fertility, nutrient availability and maintenance of humidity in soil improved morphological traits. The favorable effects of organic fertilizer on morphological traits have been reported for *Dracocephalum moldavica* L. (Hussein *et al*., 2006), *Plantago arenaria* (Hendawy, 2008), *Matricaria chamomilla* (Hadi *et al*., 2011), *Chrysanthemum morifolium* (Verma *et al*., 2011) and Marigold (Tagetes) (Paul and Bhattacharya, 2012).

As shown in Table 6, harvesting time in flowering stage (H3) had high values of stem diameter, number of secondary branches, number of flowering branches and dry weight. In addition, the lowest amount of mention traits was recorded for pre-flowering stage (H1). Concerning the effect of harvest time, in the full flowering stage was gained the greatest value of morphological traits with passage of time and it might be due to the increment of growth and development in plant tissues. Zawislak (2011) stated that morphological traits in Hyssop significantly affected by harvesting time and the largest value plant diameter and number of main shoots was recorded in full blooming, and after flowering phases respectively. As well as, they mentioned from plants harvested in flowering phase relative to vegetative phase increased considerably fresh and dry weight herb. These results are in agreement with Badi *et al*. (2004) that, reported maximum dry weigh was obtained at the beginning of flowering stage in thyme.

**Essential Oil Content and Yield**

Our finding showed that percentage of essential oil ranged for 0.474 to 0.585. According by Valtcho *et al*. (2012) essential oil content ranged from 0.13 to 0.26% while Khazaie *et al*. (2008) found that percentage of essential oil changed for 0.83 to 1.2%. Variation of oil content may be attributed to genetic differences, geographic origin, ecological factors and climatic conditions (Argyropoulou *et al*., 2007; Aziz *et al*., 2010). Application of F4 and F3 treatments gave the highest essential oil content 0.585 and 0.573% respectively while Control and chemical treatments produced the lowest essential oil content with 0.474 and 0.518% respectively. The positive effect of vermicompost and compost on essential oil percent were observed many medicinal and aromatic plants such as *Plantago arenaria* (Hendawy, 2008), *Majorana hortensis* (Hadi *et al*., 2011), *Pimpinella anisum* (Darzi *et al*., 2012), dragonhead (Hussein *et al*., 2006) and *Mentha piperita* L. (Ayyobi *et al*., 2014). It seems that increase essential oil content in Hyssop plant treated with vermicompost attributed to improvement of soil edaphic condition, acceleration of metabolic reactions as well as enhanced availability of nutrients which encourage oil accumulation in oil-gland cells. The blooming and full flowering stages (H2 and H3) significantly improved essential oil content compare to pre-flowering stage (H1) while do not any difference between H2 and H3 phases. Our result showed that essential oil content changed during plant growing and reached to highest value in full flowering stage. Zawislak (2011) demonstrated that maximum and minimum oil content of Hyssop was found in full blooming (1.65%) and vegetative phases (0.67%) respectively. According by Hamrouni *et al*. (2009) in (*Origanum majorana* L.) percentage of essential oil for 0.04% in late vegetative stage reached to 0.09% in full flowering stage. These results are in agreement with most of previous study that, reported the full-flowering stage is characterized by the highest essential oil yield such as Ozguven and Tansi (1998) in *Thymus vulgaris*, Rohloff *et al*. (2005) in peppermint, Sefidkon *et al*. (2007) in *Satureja rechingeri* and Verdian-rizi (2008) in *Laurus nobilis*. These finding showed that production of essence is low during vegetative phase that similar finding reported by Hamrouni *et al*. (2009) in (*Origanum majorana* L.).

The same as essential oil content, using of F4 treatment (15 ton $ha^{-1}$) produced the highest essential oil yield, but it had not any significant differences with F3 (10 ton $ha^{-1}$) treatment (Table 7). In addition, application chemical fertilizers (F5) significantly reduced essential oil yield compared to F4 and F3 treatments. Also, the full flowering stage (H3) was the best in essential oil yield while pre-flowering stage produced the lowest essential oil yield.

In fact, for plant grown F4 and F3 treatments essential oil yield was high due to high value of dry weight and essential oil percent. The increase in essential oil yield for F4 and F3 treatments can possibly be attributed to the better supply of nutrients and improved soil condition by using of vermicompost. This result showed that using vermicompost in high levels with increment of photosynthesis in Hyssop could be increased synthesis and accumulation essential oil in plant tissues.

Several investigators mentioned similar results on different plants such as *Pimpinella anisum* (Darzi *et al*., 2012), dragonhead (Hussein *et al*., 2006), *Mentha piperita* L. (Ayyobi *et al*., 2014) and *Plantago arenaria* (Hendawy, 2008). Our finding showed that in Hyssop plant with advancement in the time of harvesting increased essential oil yield due to improvement in dry mass accumulation and essential oil percent. Similar to our finding earlier researchers such as Badi *et al*. (2004) and Valtcho *et al*. (2012) reported that in species belonging to the *Lamiaceae* family mainly *Thymus vulgaris* L. and *Hyptis suaveolens*, the greatest essential oil yield was gained during the flowering stage. Also, it seems that selecting suitable harvesting time is very important for reach maximum essential oil yield.

**Table 7** Mean comparison of the measured photosynthesis pigments of hyssop (*Hyssopus officinalis* L.) for vermicompost and chemical fertilizer

| Fertilizer | CA mg $g^{-1}$ FW | CB mg $g^{-1}$ FW | TC mg $g^{-1}$ FW | C mg $g^{-1}$ FW | TF mmol$g^{-1}$FW |
|---|---|---|---|---|---|
| F1 | $0.716^{c}$ | $0.471^{b}$ | $1.18^{c}$ | $0.153^{c}$ | $355.11^{a}$ |
| F2 | $0.821^{bc}$ | $0.556^{ab}$ | $1.37^{bc}$ | $0.184^{abc}$ | $378.46^{a}$ |
| F3 | $0.843^{bc}$ | $0.521^{b}$ | $1.36^{bc}$ | $0.193^{ab}$ | $388.37^{a}$ |
| F4 | $1.030^{a}$ | $0.628^{a}$ | $1.66^{a}$ | $0.209^{a}$ | $386.51^{a}$ |
| F5 | $0.957^{ab}$ | $0.564^{ab}$ | $1.52^{ab}$ | $0.161^{bc}$ | $309.22^{b}$ |

Traits are: chlorophyll a (CA), chlorophyll b (CB), total chlorophyll (TC), carotenoid (C), and total flavonoids (TF). Fertilizer: F1: control, F2: 5 ton $ha^{-1}$, F3: 10 ton $ha^{-1}$, F4: 15 ton $ha^{-1}$ and F5: urea or N 75 kg $ha^{-1}$. The same letters in each column indicate that there is no significant difference between the means

**Table 8** Mean comparison of the photosynthesis pigments of hyssop (*Hyssopus officinalis* L.) for three levels of harvesting time

| Harvesting | CA mg $g^{-1}$ FW | CB mg $g^{-1}$ FW | TC mg $g^{-1}$ FW | C mg $g^{-1}$ FW | TF mmol$g^{-1}$FW |
|---|---|---|---|---|---|
| H1 | $0.783^{b}$ | $0.479^{b}$ | $9.26^{b}$ | $0.117^{a}$ | $337.21^{b}$ |
| H2 | $0.945^{a}$ | $0.571^{a}$ | $1.51^{a}$ | $0.185^{a}$ | $385.08^{ab}$ |
| H3 | $0.897^{ab}$ | $0.594^{a}$ | $1.49^{a}$ | $0.178^{a}$ | $386.32^{a}$ |

Traits are: chlorophyll a (CA), chlorophyll b (CB), total chlorophyll (TC), carotenoid (C) and total flavonoids (TF). Harvest time: H1: pre-flowering, H2: blooming and H3: full flowering. The same letters in each column indicate that there is no significant difference between the means

**Chlorophyll and Carotenoid**

As shown in Table 7, the forth level of fertilizer regime or F4 (15 ton $ha^{-1}$) had high values of chlorophyll (a, b and total) whereas, it had not any significant differences with F5 treatment. Control treatment gave the lowest chlorophyll content. The maximum carotenoid content (0.209 mg $g^{-1}$ FW) was obtained using the F4 treatment, whereas the minimum carotenoid (0.153 mg $g^{-1}$ FW) was obtained using the F1 (control) treatment. The collected plant in blooming and full flowering (H2 and H3) stages produced the greatest chlorophyll contents but had not significant difference between H2 and H3 phases. Further, in pre-flowering stage were obtained the lowest contents of chlorophyll and total carotenoid.

According by Zawislak (2011) harvesting time had considerable effect on chlorophyll and carotenoid content in Hyssop and the maximum and minimum amount of these pigments was recorded in beginning and after of flowering stages respectively. Data obtained indicated that using of vermicompost up to 15 ton $ha^{-1}$ resulted in increasing chlorophyll a, b, total and carotenoid. This finding might be reasonable because vermicompost with positive effect on growth traits in plant leads to accumulation of photosynthetic pigments that were previously recorded by Ayyobi *et al*. (2014) in peppermint, Befrozfar *et al*. (2013) in (*Ocimum basilicum* L.), Amin Salehi *et al*. (2016) in (*Matricaria chamomilla* L.) and Vineeta *et al*. (2015) in (*Tagetes minuta* L.).

It seems that adding maximum level of vermicompost in soil due to gradually release of nutriments especially nitrogen and reduction of nitrogen leaching increased uptake N which resulted high values of chlorophyll and carotenoid contents for Hyssop plant. Similar results stated that by Amin Salehi *et al*. (2016) in (*Matricaria chamomilla* L.). The necessary of components in chlorophyll molecule are nitrogen and magnesium. Increment of availability plant root to N and Mg cause to enhanced chlorophyll concentration and leading to better photosynthesis activity and yield production. Further, may be organic manure application can be increment of chlorophyll content and photosynthesis activity due to the favorable role of N and Mg in production and accumulation of chlorophyll in plant. Similar report has been showed by (Amujoyegbe *et al*., 2007).

**Total Flavonoids**

The total flavonoids amount was significantly reduced in plants that grown F5 treatment and no significantly difference among other treatments (F1, F2, F3 and F4). The cutting plants in the full and blooming stages (H2 and H3), produced the highest amount of flavonoids and the minimum of flavonoids was obtained in pre-flowering stage. Flavonoids are a huge group of phenolic secondary plant metabolites which display antioxidant and free radical scavenging activity (Van Acker *et al*., 1995; Pietta, 2000). These results are in agreement with Papageorgiou *et al*. (2008) who reported that flavonoids were predominant during the flowering stage of (*Origanum majorana* L.). Alizadeh (2011) state that the most amount of phenolic content and antioxidant activity in Hyssop herbal was obtained in flowering stage. Our result showed that application of chemical fertilizer treatment had negative effect on flavonoid content. In this regard Dahui *et al*. (2010) stated that application high nitrogen decreased the concentrations of total flavonoids by 18%–35% compared to a low N-supply rate. Other researcher showed that nitrogen application may be inhibit the synthesis of flavonoids by increasing the channeling of L-phenylalanine towards proteins (Margna *et al*., 1989). Finding of authors showed that biosynthesis of secondary metabolites such as flavonoids increased in organic farming systems relative to conventional systems (Strissel *et al*., 2005). Other finding recorded that application of vermicompost compare to compost treatment produced the maximum flavonoid and phenol contents in *Asparagus racemosus* (Saikia and Upadhyaya, 2011).

**Total Anthocyanins**

The fertilizer × harvesting time interaction showed that fourth level of fertilizer regime (F4) had high values of anthocyanin in blooming and full flowering stages (H1 and H3) but no significant difference in blooming stage (H2) in treatments (Figure 2). Our finding showed that application of vermicompost improved amount of anthocyanin in H1 and H3 stages. Other finding showed that for Strawberry plants grown in soil supplemented with compost significantly enhanced anthocyanin content in their fruit (Wang and Lin, 2003). It has been reported that anthocyanin can help to reduce damage caused by free radical activity like platelet aggregation and endothelium-dependent vasodilation of arteries (Cao *et al*., 1997; Heinonen *et al*., 1998). It seems that vermicompost in high level had proper effect on anthocyanin content and the best harvesting time for anthocyanin amount was in H1 and H3 phases. According Connor *et al*. (2002) mineral nutrient availability was one of factors that effluence on total phenolics and total anthocyanins.

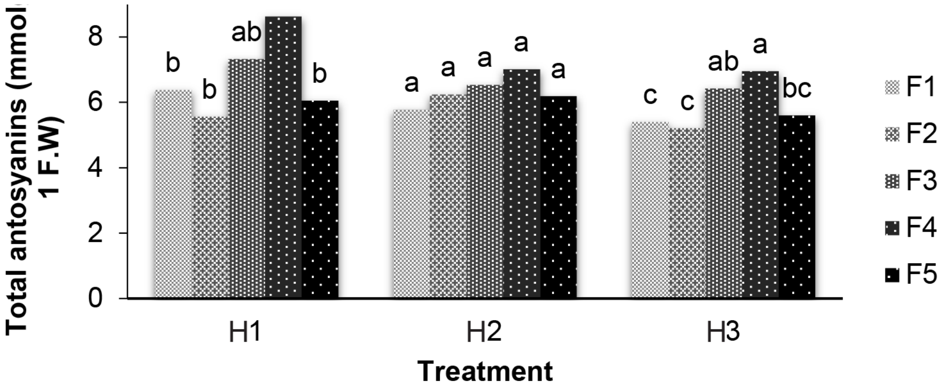


**Figure 2** The interaction between fertilizer and harvesting time on total anthocyanins of Hyssop Fertilizer: F1: control, F2: 5 ton $ha^{-1}$, F3: 10 ton $ha^{-1}$, F4: 15 ton $ha^{-1}$ and F5: urea or N 75 kg $ha^{-1}$. The same letters in each column indicate that there is no significant difference between the means. Harvest time: H1: pre-flowering, H2: blooming and H3: full flowering. The same letters in each column indicate that there is no significant difference between the means

## CONCLUSION

We determined that the all of the measured traits (exception of Carotenoid) were significantly affected by harvesting times. Moreover, full flowering was the best stage for harvesting hyssop plant and the greatest measured traits was obtained this stage particularly essential oil yield. The use of vermicompost in high level (15 ton $ha^{-1}$) improve biosynthesis and stability of biological pigments and essential oil as well as morphological characteristics in hyssop plant. The results suggest that application of vermicompost reduced the amount of chemical fertilizers application, also prevented the environment contamination from widespread application of chemical fertilizers.

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