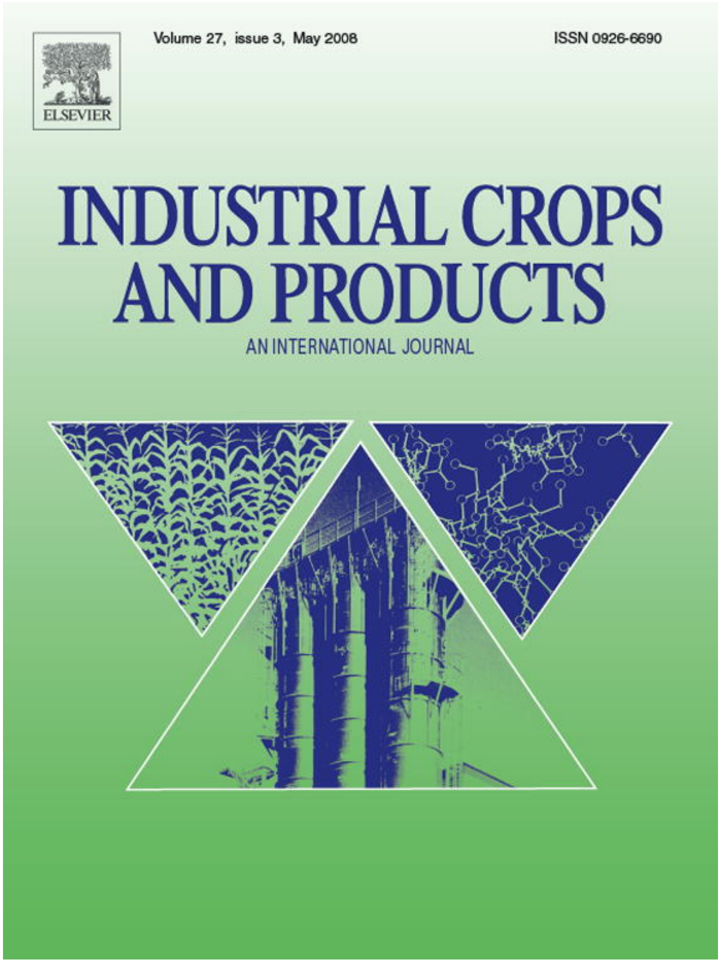
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INDUSTRIAL CROPS AND PRODUCTS

Effect of irrigation frequency and planting density on herbage biomass and oil production of thyme (*Thymus vulgaris*) and hyssop (*Hyssopus officinalis*)

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ABSTRACT

An experiment was undertaken to determine the herbage biomass and oil production of thyme (*Thymus vulgaris* Linn.) and hyssop (*Hyssopus officinalis* Linn.) in 2003 and 2004 in the semi-arid region of Khorasan in Iran. The experiment was a split plot with three irrigation intervals as main plots and three planting densities as subplots, all of which replicated three times. Irrigation intervals consisted of 7, 14, and 21 days for both crops and planting densities of 6.6, 8, and 10 (plants m^{-2}) for thyme and 5, 6.6, and 8 (plants m^{-2}) for hyssop. Different planting densities were employed by changing planting distances on cultivation rows. Herbage biomass and oil production of shoots, harvested at flowering were measured as annual production of each crop. Irrigation intervals did not change total harvested herbage biomass and oil production of both crops. Averaged across both years of the experiment, thyme produced higher oil than hyssop. Both crops produced higher biomass and oil in the second year of the experiment compared to the first year. While thyme plants biomass and oil production were lower at the highest planting density, hyssop plants showed no response to planting density. Herbage biomass and oil production of hyssop did not show a clear trend in response to interaction of irrigation intervals and planting densities in both years of the experiment. Our results showed that there is a high potential for saving water through longer irrigation intervals (e.g. 14 days) using locally adapted plants in the semi-arid conditions of Khorasan. These crops serve as alternative sources of income in dry years.

1. Introduction

To meet agricultural demands and growing competition for water, a more effective use of water in both irrigated and rainfed agriculture is essential. In regions where water scarcity is the principal limiting factor for cultivation, farmers are interested in growing crops that are able to adapt to drought conditions (Muchow, 1989; Bannayan et al., 2008). There are many farming communities within these regions which manage to survive under such conditions and, in some cases, even to succeed by exploiting natural resource bases, which their ancestors have used for generations (Reijntjes et al., 1992). Through a process of innovation and adaptation, traditional farmers have developed numerous different indigenous farming systems finely tuned to many aspects of their environment. Such risk management and adaptation strategies are in response to the limiting conditions of available water. Achieving a high crop yield, under such conditions, depends on many

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factors including the proper irrigation interval together with optimum planting density.

Crop production from late spring to the end of the summer months in the semi-arid environment of the Khorasan province in Iran mainly relies on irrigation. Shortage of water in arid and semi-arid parts of this region where annual precipitation is less than 220 mm with almost no rainfall during the summer is a prominent limiting factor of crop production. Many farmers in this study region traditionally sow local medicinal plants along with their commercial crops and believe that in dry years when they may not be able to achieve their expected harvest from their commercial crop plantation, local plants with more tolerance to drought provide an alternative source of income. In general, due to water scarcity in the study region, farmers are conservative in using the available irrigation water and try to get as much as possible from every drop. Therefore, in this region, it is of high interest to farmers to find whether a longer irrigation interval would provide an acceptable level of local crop production or not.

A better understanding of the effects of irrigation frequency and planting density on local, and neglected crops can help to determine optimal irrigation scheduling. It is expected that good management and adoption of suitable practices will improve the water conservation and result in more efficient crop production under both rainfed and irrigated conditions (Wang and Tian, 2004). Therefore, there is clearly a need to improve irrigation water management for the local farmers. However, there have been few investigations performed (Nadjafi et al., 2006; Bannayan et al., 2008) on such crops in this region. A question that also needs to be resolved is if different plant populations are relevant factors determining the final crop yield under different irrigation frequencies. In this study, we investigated the response of two local medicinal crops to irrigation frequency and planting density.

1.1. *Study crops*

There are several hundreds of underutilized and neglected medicinal and industrial crops that have been grown locally for centuries and which contribute to the financial security of the world's poorest people (Azam-Ali et al., 2001). Many of these crops are cultivated in hostile, tropical environments by small-scale farmers with little guidance on improved management practices. Because the production and improvement of most of such crops have been ignored, any attempts to improve their management rely on local knowledge and initiative. Thyme (*Thymus vulgaris*) and hyssop (*Hyssopus officinalis*) are two such local under-researched crops. The genus *Thymus* L. (Labiatae) consists of about 215 species of herbaceous perennials and subshrubs. The Mediterranean region can be described as the center of the genus (Stahl-Biskup and Saez, 2002). In *T. vulgaris*, also known as Spanish common thyme, the flowering period is between March and July. It can be found normally from sea level to 2000 m above sea level (Morales, 1986). Thyme species are commonly used as herbal tea, flavoring agents (condiment and spice) and medicinal plants (Stahl-Biskup and Saez, 2002). Among medicinal plants, thyme (*T. vulgaris*), as a perennial subshrub, is important as a crude drug in the production of plant medicines, due to its bactericidal and fungicidal effects. Its alcoholic extract has an antiseptic and expectorant effect and for this reason it is an important ingredient of cough medicines (Hornok, 1992). The dried herb of this plant is widely used as flavoring material in the acidifying of paprika, and cucumber in the canning industry. Its essential oil is also widely used by the cosmetic industry (Hornok, 1992; Haelvae and Cracker, 1998) and can be considered as new material in the food and cosmetic industry (Echeverrigaray et al., 2001; Jordan et al., 2006). *T. vulgaris* is commercially cultivated in many countries (Bernath, 1990) and has demonstrated economic benefits (Lange and Schippmann, 1997; Youdim et al., 2002). The highest oil yield and its relative concentration were obtained during the vegetative stage of growth (Jordan et al., 2006). *T. vulgaris* is adapted to alternative dry and rainfall periods (Jordan et al., 2006) and reasonably survives arid conditions (Hornok, 1992).

Hyssop (*H. officinalis*) is a perennial subshrub belonging to family Lamiaceae. Its dried flowering shoots are used in tea blends for cough relief, for antispasmodic effects, and to relieve catarrh. The bitter taste of the drug allows it to be used as a spice in households and in the food industry for the flavoring of meats and sauces. The hydrodistilled essential oil from the shoot portion is widely used in the food, pharmaceutical, and cosmetics industries (Hornok, 1992; Garg et al., 1999). The oil obtained from *H. officinalis* ssp. *officinalis* grown as an annual crop is colorless and possesses a sweet camphoraceaous odor (Garg et al., 1999). This plant is a typical xerophyte and is well adapted to drought and low input conditions (Hornok, 1992; Craker and Dinda, 1998).

2. Materials and methods

2.1. *Study site*

This study was conducted in the 2003 and 2004 growing seasons at the experiment station of the College of Agriculture, Ferdowsi University of Mashhad (latitude: 36°15′N, longitude: 59°28′E: 928 m) in the central part of Khorasan province, Iran. Climate in this province varies from semi-dry and locally humid in the north to dry in the southern parts. The Khorasan Province rainfall regime is characterized by large inter-annual variability. Annual rainfall in the north of Khorasan province is about 250 mm while in the south is 110 mm. Precipitation falls in winter as snow on the mountains of the north and west and rainfall mainly occurs between November to May. Annual mean temperature in the north of the province is about 13 °C and in the southern part is 18 °C. Maximum and minimum temperatures and precipitation of both years of this experiment are shown in Fig. 1. The soil of the field was silty loam (fine-loamy, mixed, mesic, calcixerollic, xerochrepts) with pH 7.8, contains total N (703 ppm), total P (2 ppm), and total K (0.23 ppm) with an EC of 10 mmohs cm^{-1}.

2.2. *Experimental design*

A split plot experiment based on a randomized complete block design with three replications was used. Main plots (45 m^2) were irrigated up to field capacity (26.5% θ) with 1.5 cm of water (675 L) in each irrigation. Furrows in each plot were irrigated by a hose (4 cm diameter) with a counter on it. Irrigation intervals

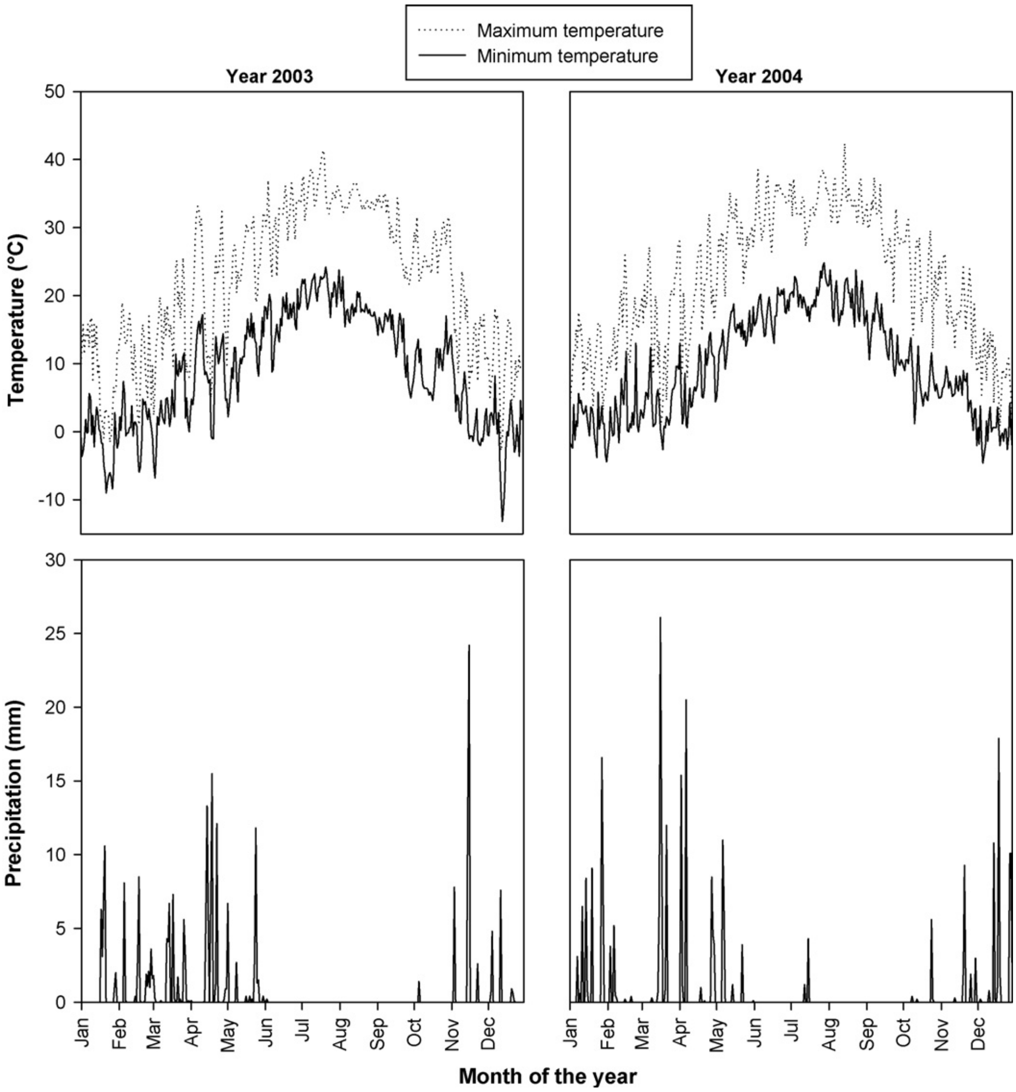

Fig. 1 – Daily values of maximum and minimum air temperature (°C) and precipitation (mm) of years 2003 and 2004.

including of 7, 14, and 21 days were arranged in main plots and three planting distances including of 20, 25, and 30 cm for thyme and 20, 30, and 40 cm for hyssop were employed in subplots. Such distances provided 10, 8, and 6.6 plants m^{-2} for thyme and 10, 6.6, and 5 plants m^{-2} for hyssop. Thyme plants were propagated on 26 March 2003, from a 3-year-old plant by root divisions. The seeds of locally bred hyssop, were sown directly in the field on 29 April 2003. All plots were fertilized uniformly with cow manure at 30 tonnes ha^{-1}, in the autumn of each year. Each experimental subplot was arranged by 6 m long and 2.5 m wide with a total area of 15 m^2. In order to prevent the lateral spread of water, plots were surrounded by dikes with a distance of 2 m between plots. Two irrigations for thyme and three for hyssop were employed with an interval of 7 days immediately after planting for uniform emergence and establishment of seedlings before starting the irrigation treatments. In the second year of the study, two irrigations were used again before starting the irrigation treatments for both plants. Weeds were controlled by hand when needed. Plants were cut from 1 m^2 area just above the lignified parts of the stem. Two cuts, in the full flowering stage, on 17 May and 20 October 2003 and two cuts on 19 May and 25 September 2004 were performed for thyme. For hyssop, one cut in full flowering stage on 22 June in 2003 and two cuts on 17 May and 4 September in full flowering stage in 2004 were performed. Collected materials were weighted after drying in a heater at 45 °C for 72 h. Essential oils were extracted from dried aerial parts of the collected samples of each treatment by hydrodistillation for 3 h, using a Clevenger-type apparatus (British Pharmacopeia, 1998). The data were analyzed by one-way ANOVA using the Statistical Analysis System (2001) and means were compared by Duncan's multiple range test at 5% probability level.

3. Results and discussion

3.1. *Irrigation interval*

Irrigation intervals, except for thyme at the 21-day interval in year 2003, did not show any effect ($P > 0.05$) on herbage biomass

Table 1 – Effects of irrigation interval on herbage biomass, oil production and oil percentage of *Thymus vulgaris* and *Hyssopus officinalis*

Irrigation interval (d)	Herbage biomass ($g\,m^{-2}$)		Essential oil yield ($g\,m^{-2}$)		Essential oil (%)	
	2003	2004	2003	2004	2003	2004
T. vulgaris						
7	1156.0	1313.7	18.9	19.6	1.6	1.4
14	1157.0	1247.5	21.2	18.7	1.7	1.4
21	726.0	992.2	13.0	16.9	1.8	1.7
LSR	170.2	ns	7.8	ns	ns	ns
H. officinalis						
7	550.2	2005.8	6.2	16.7	1.1	0.83
14	442.3	2009.1	5.4	19.1	1.2	0.99
21	320.7	1817.7	3.7	15.8	1.1	0.88
LSR	ns	ns	ns	ns	ns	ns

d, day; LSR, least significant range; ns, non-significant.

production in both years of the experiment for both crops. Essential oil production was also not different in response to irrigation interval for hyssop for the 2 years though it was significant only in the first year of the experiment for thyme (Table 1). However, herbage biomass production of both crops, in both years of the experiment was reduced as irrigation intervals increased. Generally, both species produced higher biomass and essential oil yield in the second year compared to the first year across all irrigation intervals. Averaged across all irrigation interval levels, both herbage biomass and oil production of thyme showed no significant difference ($P > 0.05$). Hyssop was not able to produce the same amount of herbage biomass and oil in the first year of experiment compared to thyme. However, the rate of increase in the second year compensated for the lower biomass and oil production of the first year. Averaged over both years of the experiment, our results showed that herbage biomass of both crops gradually decreased as irrigation intervals increased but the reduction was not significant ($P > 0.05$).

Averaged over both years, thyme oil production was significantly ($P < 0.05$) higher than hyssop at each level of irrigation. As there was not much difference between weather conditions of both years of the experiment (Fig. 1), the difference between the 2 years on production of hyssop might be due to higher efficiency of water use in the second year, probably due to a more developed rooting system. Such capability is considered as a valuable trait for a cash crop in dryland agroecosystems. There are various studies on the effects of irrigation intervals on production of different crops. Lawal and Rahman (2007) employed different irrigation intervals varying from 5 to 15 days in 2 years, and they found different yields of pepper (*Capsicum annuum* L.) in the first year of experiment. However, they reported that different irrigation intervals did not affect pepper yield in the second year of their experiment. Uçan et al. (2007) found no difference in the yield of sesame (*Sesamum indicum* L.) using irrigation intervals of 7, 14, and 21 days in a 2-year experiment. They concluded that the irrigation interval could be extended to 21 days without decreasing yield. Irrigation regimes on *Nepeta binaludensis* Jamzad, with the common Persian name of pune-sa, showed that the highest herbage biomass and essential oil yield were obtained at irrigation intervals of 7 and 14 days compared to 21 days (Nadjafi, 2006). Mirsa and Sircastava (2000) showed that moderate water stress decreased leaf area, herbage biomass and essential oil yield in peppermint (*Mentha piperita*). Katsoulas et al. (2006) studied the effect of irrigation frequency on production and quality of

Table 2 – Effects of planting density on herbage biomass, oil production and oil percentage of *Thymus vulgaris* and *Hyssopus officinalis*

Planting distance (cm)	Planting density (# m^{-2})	Herbage biomass ($g\,m^{-2}$)		Essential oil yield ($g\,m^{-2}$)		Essential oil (%)	
		2003	2004	2003	2004	2003	2004
T. vulgaris							
20	6.6	1110.0	1217.2	20.9	20.6	1.8	1.6
25	8	1122.0	1212.0	20.3	17.9	1.8	1.4
30	10	850.6	1124.3	12.0	16.7	1.6	1.5
LSR		126.0	ns	5.7	ns	ns	ns
H. officinalis							
20	5	448.9	2095.8	5.3	17.6	1.2	0.87
30	6.6	418.5	1850.8	4.7	16.1	1.1	0.88
40	8	446.3	1885.8	5.3	17.8	1.2	0.95
LSR		ns	ns	ns	ns	ns	ns

#, number; LSR, least significant range; ns, non-significant.

rose (*Rosa hybrida*, cv. First Red) flowers and found that higher irrigation frequency improved the biomass production but did not affect the quality of harvested flowers.

Our results showed that irrigation intervals had no effect (Table 1) on essential oil percentage. Both species showed a lower oil percentage in the second year compared to the first year. For thyme the highest oil percentage was obtained at the 21-day irrigation interval, but for hyssop it was obtained at the 14-day irrigation interval (Table 1). Simon et al. (1992) reported that water stress increased essential oil amount of sweet basil (*Ocimum basilicum*). Baher et al. (2002) showed that severe water stress (66% field capacity), decreased yield of savory (*Satureja hortensis*) but essential oil percentage was increased. Karamzadeh (2003) also reported that water stress increased the essential oil percentages of lavender (*Lavandula officinalis*) and absinthium (*Artemisia absinthium*).

3.2. *Planting density*

Optimum planting density is a key to achieve maximum crop production especially when water is a limiting factor. Our results (Table 2) showed that in 2003 for thyme the only significantly ($P > 0.05$) difference for herbage biomass was obtained at the highest planting density (10 plants m^{-2}) compared to the two other planting densities (8 and 6.6 plants m^{-2}). In 2004, there was no significant ($P > 0.05$) effect of density on herbage biomass. For hyssop different planting density did not show any effect on herbage biomass production in both years of the experiment. Comparison of the two plants herbage biomass production in response to different densities in 2 years showed that thyme in its second year was able to tolerate the density pressure.

Oil production (Table 2) of thyme followed a similar trend as its herbage biomass production in both years. The highest oil production (20.9 g m^{-2}) was obtained in 2003 at lowest planting density (6.6 plants m^{-2}) while the lowest oil production (12 g m^{-2}) was obtained in the same year at the highest planting density (10 plants m^{-2}). However, oil percentage showed no difference in both years (Table 2). For hyssop different planting densities did not result in change of amount of oil and percentage of oil in both years (Table 2). The highest oil production (17.8 g m^{-2}) was obtained in 2004 at the 40 cm planting distance (8 plants m^{-2}) and the lowest (4.7 g m^{-2}) was obtained in 2003 at the 20 cm planting distance (5 plants m^{-2}). Comparing the two species, the herbage biomass was similar but oil production of thyme was higher than for hyssop. Plant population density has important effects on vegetative development of maize (Tetio-Kagho and Gardner, 1988). If plant density is too high, the decrease in the availability of water per plant generates a marked fall in yield per plant that is not offset by the increase in the number of plants (Vega et al., 2001). Shalaby and Razin (1992) reported that herbage biomass and essential oil production of thyme increased at lower planting distances. El-Gendy et al. (2001) showed that the lowest planting distance (15 cm) resulted in higher biomass and essential oil yield compared to a 45 cm planting distance in sweet basil (*O. basilicum*). Nadjafi (2006) reported that the highest herbage biomass and essential oil yield of pune-sa (*N. binaludensis*) was observed at a planting distance of 25 cm, compared to 50 cm. Non-significant differences of herbage biomass and oil production of thyme between different planting distances in the second year indicated that individual plants of this species produced more branches and shoot growth at a lower planting distance to compensate for the lower number of plants per unit. Essential oil content was not affected by the planting distance which was also reported by other investigators (Shalaby and Razin, 1992; El-Gendy et al., 2001; Nadjafi, 2006). Hossein et al. (2006) showed that wider plant distances of dragonhead (*Dracocephalum moldavica*) increased the herbage biomass. They mentioned that individual plants in wider distances had more branches and herbage yield compared to the narrow distances. Planting distance showed an effect on essential oil percentage, which was reported also for other plants such as pune-sa (*N. binaludensis*) (Nadjafi, 2006), *T. vulgaris* (Shalaby and Razin, 1992), and common basil (*O. basilicum*) (El-Gendy et al., 2001).

3.3. *Interactive effects of irrigation and planting density*

Increasing the irrigation interval from 7 to 14 days showed the highest interaction of the two treatments on herbage biomass at the lowest planting density (6.6 plants m^{-2}) for thyme in 2003 (Table 3). Similar results were obtained when irrigation interval increased from 7 to 21 days (Table 3). In 2004, the extent of herbage biomass reduction due to longer irrigation interval, from 7 to 14 days and from 7 to 21 days, was lower as planting density increased. Interactions of planting densities and irrigation intervals in both years showed a similar direction as interactive effect on herbage biomass.

For hyssop plants, increasing the irrigation interval from 7 to 14 days resulted in a higher production only at the planting distance of 30 cm (6.6 plants m^{-2}) in the first year of the experiment. Increasing the irrigation interval from 7 to 21 days generally resulted in lower production in both years compared to 14 days (Table 3). It seems when plants are older they are able to suppress the effects of water shortage and even produce more oil at longer irrigation intervals. Our observations indicated that 2-year-old plants have more herbage compared to the 1-year-old plants but differences were not significant for essential oil yield. Omidbeigi (1998) reported that increasing plant age in *T. vulgaris* resulted in higher yield and essential oil yield. The highest herbage yield (5.8 tonnes ha^{-1}) and essential oil yield (50.5 kg ha^{-1}) were obtained in 3-year-old plants compared to 2- and 1-year-old plants. Nadjafi (2006) also reported that 2-year-old *N. binaludensis* plants produced higher yield and essential oil compared to 1-year-old plants.

As expected, increasing the irrigation interval will result in less available water in the soil (Hutton et al., 2007). However, depth of rooting systems and their ability to absorb water would prominently affect the plants access to available water. We concluded that for 1-year-old plants, an irrigation frequency of 14-days would be sufficient. As plants grow older, probably due to more and deeper roots system, a longer irrigation interval of 21-days may not affect crop production of herbage biomass and oil production. The most important result to emerge from this study is that substantial saving in water may be achieved by increasing the irrigation interval and by employing local crops, which are adapted to local environments and are able to maintain both herbage biomass and oil production, especially as they get older.

Table 3 – Interactive effects of planting density and irrigation interval on herbage biomass and oil production of *Thymus vulgaris* and *Hyssopus officinalis* plants in 2 years of experiment

Planting distance (cm)	Planting density (# m^{-2})	Herbage biomass ($g\,m^{-2}$)					Oil yield ($g\,m^{-2}$)				
		Irrigation interval (d)			Ratio		Irrigation interval (d)			Ratio	
		7	14	21	14/7	21/7	7	14	21	14/7	21/7
T. vulgaris											
2003											
20	6.6	870.0	1420.0	1041.0	1.6	1.2	16.0	31.8	14.8	2.0	0.9
25	8	1297.0	1400.0	670.0	1.1	0.5	23.8	21.8	15.3	0.9	0.6
30	10	1300.0	650.0	466.7	0.5	0.4	16.9	10.1	9.0	0.6	0.5
2004											
20	6.6	1487.0	1065.3	1099.3	0.7	0.7	24.0	17.4	20.3	0.7	0.8
25	8	1393.6	1291.0	951.3	0.9	0.7	19.6	18.2	15.9	0.9	0.8
30	10	1060.6	1386.3	926.0	1.3	0.9	15.4	20.5	14.4	1.3	0.9
H. officinalis											
2003											
20	5	625.6	416.5	304.5	0.7	0.5	7.1	4.8	3.9	0.7	0.5
30	6.6	441.6	502.5	310.0	1.1	0.7	4.8	5.9	3.5	1.2	0.7
40	8	583.3	408.0	347.6	0.7	0.6	6.8	5.4	3.8	0.8	0.6
2004											
20	5	1946.6	2412.6	1928.0	1.2	1.0	15.7	20.5	16.7	1.3	1.1
30	6.6	2191.6	1736.0	1625.0	0.8	0.7	17.9	16.0	14.6	0.9	0.8
40	8	1879.3	1878.0	1900.0	1.0	1.0	16.6	20.7	16.1	1.2	1.0

d, day; #, number.

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