The influence of nitrogen-fertilizer and harvest time on the productivity of *Thymus vulgaris* L.

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Key words: Lamiaceae, *Thymus vulgaris*, garden thyme, nitrogen-fertilizer, harvest time, dry-matter production, herb yield, essential oil yield, oil composition, thymol.

Summary: The influence of nitrogen-fertilizer applied in 0, 50, 100 and 150 kg/ha dosages, as well as the time of the harvest carried out in full flowering and early fruit set stages were studied on the herb and essential oil production of garden thyme *(Thymus vulgaris* L.). The small plot experiment was installed in the Experimental Field of Tarbiat Modarres University near Teheran, under sandy loam soil conditions. On the basis of the results the nitrogen-fertilizer had a significant effect on the dry-matter production of the species: the herb yield, calculated on one hectare, increased from 671.88 kg up to 1021.00 kg value as a result of 150 kg nitrogen dosage. The essential oil yield proved to have a similar tendency because neither the accumulation level of essential oil, nor the ratio of thymol were effected by the nutrient supply. Analyzing the effect of harvest time changes in both dry-mass production and essential oil accumulation were observed. The highest herb yield (1238.20 kg/hectare) was obtained in early fruit set, when about 50 per cent of fruits reached their full size in the inflorescence. The accumulation level of essential oil also reached its maximum at the same development stage, showing 0.75 per cent value, which is about two fold higher comparing to the accumulation level was measured at the time of full flowering (0.41 %).

Introduction

According to the literature data garden thyme (*Thymus vulgaris L.)* was applied for curing even in ancient time. The drugs of the species, which are known as *Thymi vulgaris herba* and *Aetheroleum thymi* were widely utilized back to the historical times, and have been authorized since the sixteenth century (*Hornok*, 1992). The drugs of the species are present in many up to date official descriptions, including Hungarian (Ph.Hg. VII.), Swiss (Ph.Helv. VII.), Austrian (ÖAB) and German (DAB 10) Pharmacopoeias (*Anonymous*, 1984). Both of the above mentioned drugs are extensively used in phyto- and aromatherapy. Garden thyme is regarded as one of the most important elements of drug mixtures and tinctures because of its antiseptic properties. The plant is effective in treating whooping cough as well as the parasitic infections. It is used for its mouthwash value in appropriate preparations such as liquid dentifrices.

The garden thyme is a perennial dwarf shrub belonging to the *Lamiaceae* family (*Bernáth*, 1993a). It grows up to 40-50 cm in height, forming many branches. The sessile leaves vary in shape from elliptic to linear or diamond-shaped towards the apex. The flowers changhing in colours from white to purple are united in spikes at the top of the branches. The fruits consist of a smooth, dark-colored 4-sectioned nutlet found in the remains of the calyx.

The garden thyme grows wild in the Mediterranean area with the preference for a sunny position in dry, gravelly soils. In many European countries, including Spain, France, Hungary, Bulgaria etc. the plant is cultivated on a middle scale (*Bernáth*, 1993a).

Effect of environmental factors on the production of medicinal and aromatic plants (*Bernáth*, 1993b), especially on garden thyme have been studied by many authors (*Basker & Putievsky*, 1978, *McGimpsey* et al. 1994, *Rometsch*, 1993). Furthermore, it was stated that the essential oil and thymol content of thyme were dependent on both nitrogen supply as well as the stage of development. In the latter case the appearance of flowers and the actual stage of fruit development seems to have a main importance (*Ceylan* et al. 1994, *Omidbaigi*, 1998) .

Since garden thyme is cultivated commercially, the optimum amount of fertilizers and time of harvest have been the subject of many investigations (*Ceylan* et al. 1994, *Shalaby & Razin*, 1992). Our aim was to determine, whether the above mentioned environmental factors (nitrogen fertilizer and harvest time) could change the herb and essential oil production of the species cultivated under Iranian conditions.

Material and methods

Plant material

The seeds of *Thymus vulgaris* were provided by Firma Bornträger und Schlemmer GmbH from Germany, in 1994. The seeds were sown in the spring of 1994 and the plant material has been selected afterwards in the course of two vegetation cycles (1995–1996). For the experiment individuals of homogenous character were selected and propagated vegetatively in 1997. The planting was made on 5th of March, 1997.

Place and conditions of experiment

The investigation was carried out in Experimental Station of Paykan Shahr belonging to the College of Agriculture of Tarbiat Modarres University, near Tehran. The Station is situated in 1215 m above the see level. The weather condition of the Station can be characterized by semi-dry climate with 242.7 mm annual precipitation. The average humidity of the region is 42 %, while the lowest temperature is around –7 °C The main soil characteristics of the Experimental Station are as follows: sand = 75%, clay =13 %, pH = 7.9, K(ppm) = 305, P(ppm) = 4.2, Total N = 0.05%, organic C = 0.92%.

The effect of nitrogen-fertilizer and the time of the harvest were studied in two factorial experiment using randomized block design with three replications in 1997. In the case of fertilization an untreated control and three different nitrogen dosages, 50 kg, 100 kg and 150 kg nitrogen calculated to one hectare were used in form of urea. The plant pots treated with different nitrogen dosages, as well as the control ones were divided into two parts and harvested in different stages of development. These development stages were as follows: a) flowering stage, when 50 per cent of plants reached the full flowering status, b) early fruit development stage, when 50 per cent of fruits reached their full size in inflorescence. The time of the harvest was 30th of May at the first time, while 25 the June in the case of the second.

The size of each plot was 1.60 x 1,60 m containing 32 individual plants, propagated vegetatively. The distance between pots was 0.80 m, while 1.5 m between blocks. Hoeing and mechanical weeding, as well as irrigation were done regularly. The nitrogen-fertilization dosages – according to the experimental design – were added in two portions, 50 per cent on the 10th day, while second part 30 days after plantation.

Analysis of essential oil

The air- dried parts of garden thyme were distilled in Clevenger apparatus according to the method of Hungarian Pharmacopoeia (16). The quality of the essential oil, especially its thymol content, was determined by GC (gas chromatograpfhy) using Varian 4500 FID (flame ionization detector) gas chromatograph (Phillips). Separation took place on a 2m x 3 mm column packed with 3% OV-17 (80–100 mesh). Column temperature was programmed as follows: 2 min. at 60 °C at a rate of 8 °C/min., 195 °C was kept for 5 min. Injector temperature was 250 °C, the type of carrier gas was N. Thymol of 98% purity was used as a standard.

Biometrical analysis

The results were analyzed by two factorial biometrical method using Statgraph 5.0 program, installed on Personal Computer.

Results and discussion

Evaluating the results it became obvious that between the nitrogen-fertilizer and harvest time no any interaction was found, neither on herb production, nor on the accumulation and quality of the essential oil. This is the reason why the effect of the two above mentioned factors are discussed separately.

Effect of nitrogen-fertilizer

According to the results the different amount of nitrogen-fertilizer had a significant effect on the herb production of garden thyme (*Table 1*). Especially the effectiveness of 100 kg and 150 kg dosages, calculated on one hectare, has been proved. The yield of the pots fertilized by 150 kg nitrogen was 260 g, which is 50 per cent higher comparing to the control. It does mean that applying intensive nutrition 1021.00 kg herb can be produced under Iranian conditions, calculated to one hectare.

In accordance with the results of *Shalaby & Razin* (1992), *Ceylan* et al. (1994) the nutrition had no significant effect on the accumulation level of essential oil. Its value ranged about 0.60 per cent (*Table 1*). In spite of the remarkable production differences observed in dry mass production the did not have had no any effect on the thymol ratio of the essential oil. This results is in harmony with the report of *Ceylan* et al (1994). The ratio of thymol changed between 34.04 and 41.13% values, but this seems to be determined by genotype of the population (*Németh* et al. 1993., *Németh & Bernáth*, 1994).

The nutrition, in spite of unchanged accumulation level of essential oil, has a significant effect on the essential oil yield (*Table 1*). It is due to the increase of dry-matter

Table 1 Effect of nirtogen-fertilizer on herb yield, essential oil and thymol content of *Thymus vulgaris*

No.	N- fertilizer (kg/ha)	Herb yield (%)	Essential oil (g/plot)	Essential oil yield (g/plot)	Thymol (%)
1	0	170.0^{B}	0.60^{A}	1.02^{B}	34.04^{A}
2	50	100.0^{AB}	0.52^{A}	0.52^{B}	41.13^{A}
3	100	210.0^{AB}	0.59^{A}	1.24^{AB}	35.50^{A}
4	150	260.0^{A}	0.61^{A}	1.58^{A}	38.10^{A}
Duncan's test		Level 1%	Level 5%	Level 1%	Level 5%

Means followed by the similar letters in each column – according to Duncan's multiple range test – are not different significantly

production which is going on according to the linear equation type of primary and secundary compound accumulation, described by *Bernáth* (1986, 1990, 1992, 1993a). Plants receiving 150 kg/ha nitrogen-fertilizer produced the highest essential oil yield (1.58 g/pot), which is beyond the control value (1.02 g/parcel) by 50 per cent.

Effect of time of harvest

As it is demonstrated by the data shown in the *Table 2* the time of the harvest has a significant effect on herb yield of thyme. The highest yield (316.9 g/pot) was obtained in fruit set development stage, when about 50 per cent of the fruits reached their full size in the inflorescence. This yield refers to 1238.20 kg dry herb mass, calculated on one hectare.

The accumulation level of essential oil reached its maximum at the same time, in early fruit set development stage, showing 0.75 per cent value, which is about two fold higher comparing to the accumulation level measured at full flowering (0.41%). This results are in harmony by the statements of *McGimpsey et al* (1994), *Basker & Putievsky* (1978), but in contrast to the Central-European observations (*Hornok*, 1992, *Bernáth*, 1993 a). However, the time of the harvest had no effect on the thymol ratio of the essential oil, which seems to justify our earlier statement, that the quality of the oil is mainly determined by the genotype of the population.

The time of the harvest, regulating both the dry-mass production as well as the accumulation level of essential oil has a remarkable effect on the essential oil yield (*Table 2*). Due to the increase of dry-matter production and oil accumulation level plants harvested in early fruit set development stage produced as much as 2.37 g oil per pots, which is about five times higher comparing to the control value (0.45 g/pot).

Table 2 Effect of harvest time on herb yield, essential oil and thymol content of Thymus vulgaris

No.	Time of harvest	Herb yield (g/plot)	Essential oil (%)	Essential oil yield (g/plot)	Thymol (%)
1	Full flowering	110.0 ^{B}	0.41 ^{B}	0.45^{B}	38.28^{A}
2	Early fruit set	316.9 ^{A}	0.75 ^{A}	2.37^{A}	36.11^{A}
Duncan's test		Level 5 %	Level 1%	Level 1%	Level 1%

Means followed by the same letters in each column according to Duncan's multiple range test – are not different significantly

Conclusion

To conclude our result it seems to be obvious that no interaction could be found between the effect of nitrogen supply and the time of the harvest of *Thymus vulgaris*. The nitrogen-fertilizer had significant effect on the herb yield, but did not change either the essential oil accumulation level nor the quality of the oil, characterized by thymol content (*Fig. 1* and *Fig. 2.*). It does mean that both the herb and essential oil yield can be increased by nutrition. Under Iranian condition the application of 150 kg/ha nitrogen dosage proved to be the optimum one.

According to data harvest time had significant effect on both the herb yield and essential oil accumulation level, but did not change the oil quality *(Fig. 1 and Fig. 2)*, Harvesting of thyme in early fruit set stage is recommended by results of this investigation.

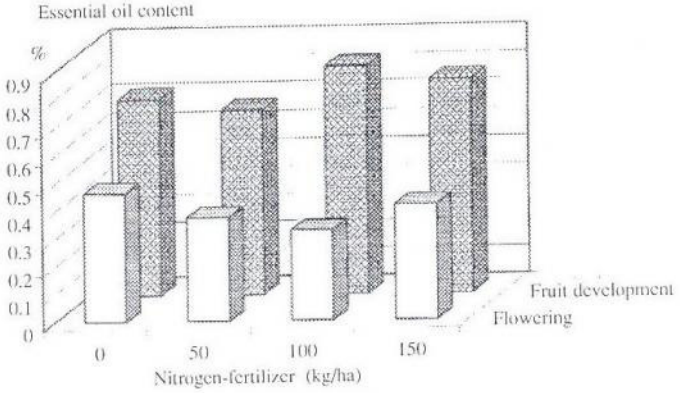

Figure 1 Effect of nitrogen-fertilizer and the time of the harvest on the essential oil accumulation of garden thyme *(Thymus vulgaris)*

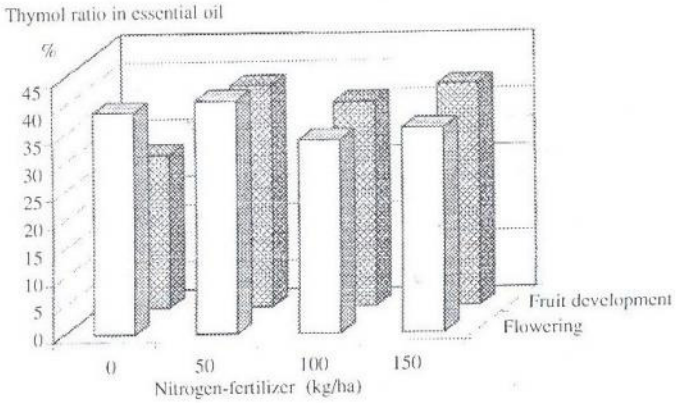

Figure 2 Effect of nitrogen-fertilizer and the time of the harvest on the thymol content of essential oil distilled from garden thyme *(Thymus vulgaris)*

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