

Nepeta Cataria L, One of the Promising Aromatic Plants In Egypt: Seed Germination, Growth and Essential Oil Production

Mohamed E. Ibrahim¹, Salma A. El-Sawi² and Faten M. Ibrahim^{1*}

¹Pharmacognosy Dept., ² Medicinal and Aromatic Research Dept., National Research Centre, 12622-Dokki, Egypt

Received 10 Sep 2016,
Revised 11 Mar 2017,
Accepted 16 Mar 2017

Keywords

- ✓ *Nepeta cataria*;
- ✓ Egyptian atmosphere;
- ✓ Soil kind;
- ✓ Germination;
- ✓ Growth;
- ✓ volatile oil;
- ✓ constituents;
- ✓ Nepetalactone.

M. E..Ibrahim,
Melsayed49@yahoo.com

Abstract

In order to add a new species of medicinal and aromatic plants to be cultivated in Egypt for export we decided to study the plant of *Nepeta cataria* L, by identifying the best ways to increase the proportion of seed germination, vegetative measurements and identify productivity of essential oil. Seeds germination of *Nepeta cataria* L plant as affected by the kind of soil germination and date of sowing were studied. Preparing seeds nursery was carried out using three different types of agricultural soil. The first one was a pure sandy soil (S1), the second was a mixture of sand and clay loamy soil (S2) in the ratio of (1: 1), while the third included a mixture of sand and clay loamy soil (S3), in the ratio of (2: 1). It is clear from the results that the soil type and sowing time treatments had significantly affected germination percentage. Using S3 treatment gave the best result particularly when the seeds were sowed during February in the two seasons. The growth measurements of *Nepeta cataria* plants during the growing season under the Egyptian environment showed that, the fresh yield of *N. cataria* herb recorded 138.5 and 180.0 gms / plant in the first and second cut, respectively. The highest percentage of oil recorded (0.25%) during the first cut, against (0.19 %) in the second cut. Analysis of *Nepeta cataria* oil indicated that, the volatile oil of *N. cataria* L from Egypt is rich in citral, geraniol, and nepetalactone. The oxygenated monoterpenes compounds were found as the main composition of essential oil of catmint oil (70.75 and 81.55 % in the first and second cut, respectively). Citral and nepetalactone were found as the main compounds of oxygenated monoterpenes group in catmint oil cultivated in Egypt.

1. Introduction

Nepeta cataria (catmint), family Lamiaceae, is native to eastern and southern Europe. It is also found in the Middle East, China and North America. The name catmint is derived from the strong attraction most cats have towards them [1-5]. *Nepeta cataria* is a perennial herb [6], it used for ornamental, cooking purposes, and as a folk-medicine. Its volatile oil used in perfumes and cosmetics industry. The essential oils content are varying during vegetation from 0.30 % to 1.2 % [7]. Nepetalactone and citral are the main constituent of the essential oil [8-10]. Seed germination is influenced by some factors such as the substrate type used and environmental factors such as temperature, light, water and oxygen [11-12]. Generally, soil kind is one of identified factors to get seedlings of high quality [13-15]. The recent work is an effort to follow the growth of catmint under Egyptian conditions starting from planting the seeds in the nursery till getting the yield of herb and volatile oil content. It aims also to add the catmint plant to the list of medicinal and aromatic plants cultivated in Egypt for export and increasing hard currency in the country.

2. Materials and methods

Seeds of *Nepeta cataria* were obtained from the ornamental plants unit in National Research Centre-Giza Egypt. An experiment was carried out during two successive seasons 2012/2013 and 2013/2014. The layout of the experiments was in complete randomized design of three replicates. The experiment included studying of seed germination using different types of soil in different dates of the year. The seeds of *Nepeta cataria* were sown every month in nurseries to study the effect of date of sowing on germination percentage. Pure sand soil (S1) and two levels of mixture of sand and clay loamy soil (S2=1 vol. sand + 1 vol. clay loamy & S3 = 2 vol. sand + 1 vol. clay loamy) were used as a germination medium. Sowing was started on the first of October, and lasted for seven months. Seedlings of *N. cataria* were planted in the field on 15th of April in hills 30 cm apart, on rows 60 cm in-between. Two cuts were taken on 15th June and 20th September in every season. The plants resulted from the propagation were compared with the mother plants (cultivated in the garden of the National

Research Center, Giza -Egypt} in terms of growth, percentage and constituents of volatile oil under Egyptian conditions

2.1. Chemical Studies:

The volatile oil from the fresh herb, were isolated by hydrodistillation for 3 hr in order to extract the essential oils according to Gunether (1961) [16]. The isolated volatile oils were dehydrated over anhydrous sodium sulfate and were stored in refrigerator until analyzed.

2.2. Analysis of Essential oils:

Samples of essential oils of *Nepeta cataria* were isolated from the fresh herb of the two cuts. Oil constituents of each sample was investigated by the means of GC and GC-MS

2.3. Gas chromatography:

FID Hewlett-Packard 5890 using a DB-5 (methyl-silicone containing 5 % phenyl groups) column 25 m x 0.31 mm i.d. Temperature program was ; 2 min at 60 °C , 60-100 °C (2°C/min) and 100-250°C (5°C/min), carrier gas was helium at flow rate of 1.0 ml/min.

2.4. Gas chromatography-Mass spectrometry:

A Hewlett packard 5989A GC-MS system equipped with library software Wiley 138 and NBS75 was used. Capillary GC conditions as mentioned above were employed for DB-5 column. Injection volume was 1.0 μ l at 1:50 split. Significant MS operating parameter: ionization voltage 70 ev, scan mass range 40-350 *m/z*.

2.5. Identification of components:

Compounds were identified by matching their mass spectra with those recorded in the MS library [17] Adam 1995) and further confirmed by injecting the authentic samples of different compounds with the volatile oil and by comparing the mass spectra with those of reference compounds or with the published data.[18] Kovats,1959

2.6. Soil analysis:

Soil physical and chemical analysis for the different germination soils will be determined by hydrometer method Piper (1950) [19], (Table 1, 2).

2.7. Statistically analyzed

The least significant difference (LSD at level of 5%) was used to compare between different means according to Snedecor and Cochran (1982) [20] .

Table 1. Mechanical analysis clay and sand soil

Soil sample	Coarse sand	Fine sand	Silt + Clay %
Clay loam	40	18	42
Sand	71	17	12

Table 2. Chemical analysis of the different germination soils

Items	S1	S2	S3
pH	8.6	7.7	7.9
Total N ppm	120	210	190
Water soluble P ppm	0.6	0.5	1.1
Water soluble K ppm	2.7	1.1	4.2
EC (dSS1)	1.2	1.1	1.4

S1= Pure sand soil

S2=1 (vol. sand + 1 vol. clay loamy)

S3 = (2 vol. sand + 1 vol. clay loamy)

3. Results and discussion

3.1. Germination percentage:

In general data in Table 3 reported that, germination percentage of *Nepeta cataria* varied between 30 to 80 % under all condition of soil type during different times. Data tabulated in the same table showed that, (S3) treatment gave the highest percentage of germination 56.86 and 62.86% for the first and second seasons, respectively. The highest percentage of germination was obtained when the seeds of *N. cataria* were sown in (1st February to 1st March) in the soil media consisted of a mixture (2 vol. sand + 1 vol. clay loamy). The germination percentage in this respect reached to 70% and 80% for the S3 treatment during February in the two seasons, respectively. In conclusion, S3 (2 vol. sand + 1 vol. clay loamy) treatment gave successful result during springs month in February and March [average temperatures range between (20 °C to 25 °C) for maximum temperature and (10 °C- 15 °C) for low temperature]. Many investigators studied the effect of different types of soil and planting date on seed germination of some medicinal and aromatic plants. It was found that the germination of seeds is affected by several factors such as soil type used, as well as environmental factors such as water, oxygen, temperature and light [11-12]. Overall, the soil type is one of the specific factors for high-quality seedlings [13-15]

Table 3. Germination percentage of catmint seeds as affected by soil type and date of sowing

Media Months	First season				Second season			
	S1	S2	S3	Mean	S1	S2	S3	Mean
Oct.	30	30	35	31.67	25	25	30	26.67
Nov.	40	42	45	42.33	30	30	50	36.67
Dec.	50	55	62	55.67	50	60	70	60.0
Jan.	60	60	65	61.67	60	60	70	63.3
Feb.	60	65	70	65.0	60	70	80	70
Mar.	55	55	65	58.33	60	60	70	63.3
Apr.	40	45	56	47.0	50	60	70	60
Mean	47.86	50.28	56.86		47.86	52.14	62.86	
L.S.D. at 0.05								
Months	3.358				3.733			
Media	2.198				2.443			
Months × Media	4.853				5.264			

3.2. Vegetative growth characters:

Growth measurements of *N. cataria* in the growing season (April- October) under the Egyptian conditions, were found in Table 4. It was observed from the data, that the mean height of the plants reached up to 47.1 cm in the first cut (15th June) and 37.5 cm in the second cut (20th September), respectively.

Table 4. Growth and oil production of catmint plant under conditions of Egypt.

Characteristics	Plant height cm/plant.	Fresh weight g/plant	Fresh weight Ton/hect.	Dry weight g/plant.	Dry weight Ton/hect.	Oil %	Oil yield ml/plant.	Oil yield L/hect.
First cut	47.1	138.5	7.62	35.7	1.98	0.25	0.347	19.03
Second cut	37.5	180.0	9.90	50.2	2.77	0.19	0.342	18.83
Total	84.6	318.5	17.52	85.9	4.75	-	0.69	37.86
Means	42.3	159.25	8.76	42.95	2.37	0.22	0.344	18.93
L.S.D. at 0.05	5.47	17.16	0.97	3.96	0.25	0.04	0.09	

The fresh weight of *N. cataria* herb recorded 138.5 and 180.0 gms / plant in the first and second harvest, respectively. On the other hand the yield of fresh weight recorded 7.62 ton / hect, in the first harvest, while it was 9.90 ton / hect. in the second harvest, respectively. The total fresh weight obtained from the two cut recorded 17.52 ton / hect. The same trend was observed with the dry weight of plant, which amounted to 35.7 and 50.2 g /

plant for the first and second harvest, respectively. The dry yield per hectare in this respect recorded 1.98 and 2.77 ton/hect. The total yield of dry weight from the two cuts was 4.75 ton / hect. in the first and second harvest, respectively. The results showed that catmint cultivated under Egyptian atmosphere conditions has given encouraging results on the growth and yield of herb, these results are similar to the results of the plants growing in other areas such as the United States [21]

3.3. Essential oil content:

The highest percentage of essential oil isolated from fresh herb in the first cut was (0.25 %) compared with the second cut (0.19 %). The yield of essential oil per plant recorded 0.35 ml / plant in the first cut and 0.34 ml/plant in the second cut, respectively. The total oil yield of the two cuts recorded 37.86 L/ hect.

By following-up the essential oil contents in different plant parts (leaves, stems, flowers and seeds) in different growth stages, Fig. (1)), it was found that the leaves of *N. cataria* gave significant amounts of essential oil especially in the flowering stage (0.33% fresh weight, 0.45 % dry weight). Moreover, it was observed that, the percentage volatile oil in the appropriate quantity in flowers. It was also observed the presence of very small traces of volatile oil in the stems in different growth stages.

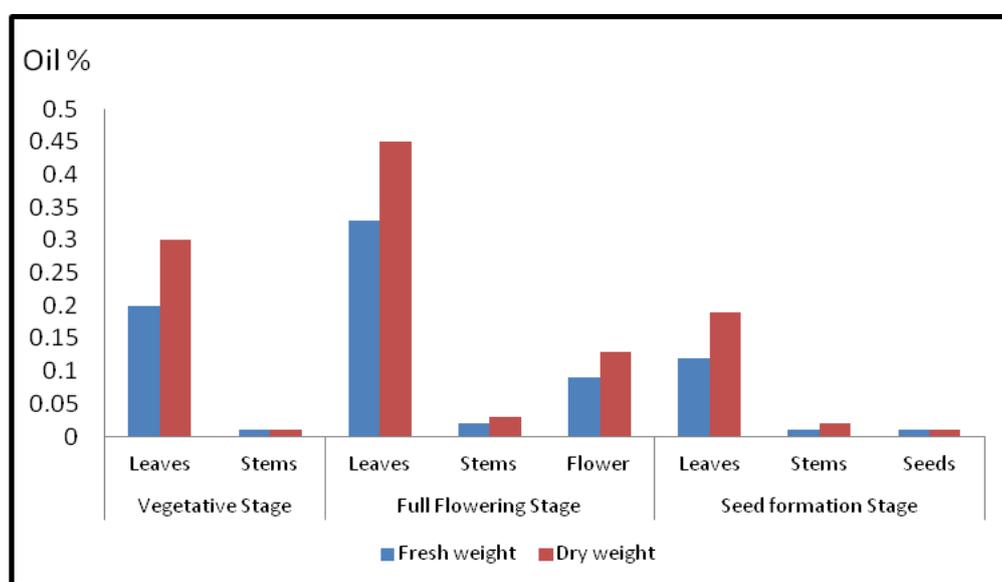


Figure 1: Volatile oil percentage in fresh and dry weight of different organs of catmint grown in Egypt

3.4. Essential oil analysis:

GC analysis of the *N. cataria* oil recorded that, eighteen components were identified; they represent about (81.88%) of the volatile fraction in the first cut, against (92.96 %) in the second cut. The fraction of monoterpene hydrocarbons, amounted to (4.31 and 4.28%), in the oil of the first and second harvest, respectively. α -Pinene, sabinene, limonene and cis ocimene were found as the main composition of catmint oil in both cuts. The values of these compounds in the catmint oil were low and with slightly different values in both cuts. In the two samples of catmint oil isolated from the herb of the first and second cut, the oxygenated monoterpenes compounds were found as the main composition of essential oil of catmint oil from Egypt. The values of this group recorded 70.75 and 81.55 % in the first and second cut, respectively. Citral, nepetalactone, geraniol, nerol, linalool, citronellol and cis verbenone were found as the main compounds of oxygenated monoterpenes group in catmint oil. The main constituents of citral (a+b) in our work recorded 31.1 % in the sample of first cut against 36.9 % in second cut. Citral has been previously recorded as the main composition of catmint oil [8], while geraniol and nerol compounds were found as the main components by Hornok *et al* [6]. Nepetalactone was found to be the main constituents of catmint oil under the conditions of Egypt. It recorded 22.6 and 24.67% in the oil sample of the first and second cut, respectively. The fraction of sesquiterpenes hydrocarbons isolated from catmint oil recorded only 3.7 and 4.2 %. It consisted of β -caryophyllene and α -humulene compounds. Sesquiterpenes oxygenated derivatives recorded only 2.12 and 1.25 % in the samples of the first and second harvest, respectively. This fraction contains germacene-D-4-ol and caryophyllene oxide compounds.

Table 5. Oil constituents of Egyptian catmint herb in the first and second cut.

Peak No	Compound	First Cut %	Second Cut %
Monoterpene hydrocarbons Compounds			
1	α -Pinene	0.700	0.750
2	Sabinene	0.613	0.550
5	Limonene	1.250	1.330
6	<i>cis</i> - Ocimene	1.750	1.650
	Total	4.313	4.280
Oxygen containing monoterpenes Compounds			
1	Linalool	0.510	0.630
2	Citronellal	0.600	0.850
3	α -Terpineol	0.371	0.500
4	Verbenone	0.612	0.350
5	Nerol	0.715	1.500
6	Citronellol	0.692	1.250
7	Neral (citral b)	13.500	14.800
8	Geraniol	13.550	14.900
9	Geranial (citral a)	17.600	22.100
10	Nepetalactone	22.600	24.670
	Total	70.750	81.550
Sesquiterpenes hydrocarbons Compounds			
11	β -Caryophyllene	2.450	2.970
12	α -Humulene	1.250	1.260
	Total	3.700	4.230
Oxygen containing Sesquiterpenes Compounds			
1	Germacrene-D-4-ol	1.900	1.660
2	Caryophyllene oxide	1.220	1.250
	Total	3.120	2.910
		81.88	92.96

Conclusion

The volatile oil of *Nepeta cataria* L under the condition of Egypt is rich in citral, geraniol, and nepetalactone. This mean that the essential oil of *N. cataria* cultivated in Egypt have two of the excellent famous aroma constituents; geraniol and citral, in addition to nepetalacton (the attractant compound), which is giving the present plant its power and activity. Also, the cultivation of the *Nepeta cataria* under Egyptian atmosphere conditions gave encouraging results and can be considered as one of the new sources of essential oils in Egypt, and increasing hard currency in the country. This requires further study of the post-harvest to produce high quality essential oils compatible with the global market.

References

1. Bourrel -C.; Perineau F.; Michel,-G.; Bessiere J.M., *J-Essent-Oil-Res-JEOR. Wheaton, Ill.: Allured Publishing Company.* 5 (1993)159.
- 2 De Pooter, H. L., Nicolai, B., De Laet, J., De Buyck L. F., Schamp N. M. and Goetghebeur., *Flavour Fragr J* (1988) 155.
3. Faten I., El Hady S., Ibrahim M.E., Youssef A.A.; Abdel Hamid M.F., *Arb. Univ. J. Agric. Sci.*12 (2004) 2.
4. Hanan F. Aly ; Ebtissam A. M.; Ibrahim M. E.; Hemaia M. M.; Faten M. I., *Journal of American Science* (2010)8.
5. Faten I., M. H. *.M.Sc.thesis (Agric. Chem.) Fac. of Agric. Ain – Shams Univ. (2005)*

6. Hornok L., Domokos J., Hethelyi E. B., *Acta-hortic. Wageningen : International Society for Horticultural Science*. May, 306, (1992) 290.
7. Dmitriev L. B., Mumladze M. G., Esvandija G. A., Granberg I. I., Jakubasvili I. Z., *Izv. TSHA* 3, (1981) 75.
8. Holubek J., Kroulik B., *Balbis Pharmaz. Zhalle*, 1, (1957) 52.
9. De Pooter H. L., Nicolai B., De Laet J., De Buyck L. F., Schamp N., Goetghebeur M., *Flavour Fragr J*, 3, (1988) 155.
10. Bourrel C., Perineau F., Michel G., Bessiere J.M., *J-Essent-Oil-Res-JEOR. Wheaton, Ill. : Allured Publishing Company*. Mar/Apr. 5,2, (1993) 159.
11. Hartmann H.T., Dale E., Kester Fred T., Davies Jr., Geneve R.L., *Plant Propagation: Principles and Practices: New Delhi, India*. (2007) 32.
12. Baiyeri K.P., Mbah B.N., *Afr. J. Biotechnol.*, 5, (2006) 1405.
13. Dickens Dolor. Am. J., *Biotechnol. Mol.Sci.*1, 25, (2011) 1.
14. Omokhua G.E., Ogu A., Oyebade B.A., *International journal of scientific & technology research* ,4 (2015) 3.
15. Süleyman G. H., Cemal G., Zafer Ö., *Afr. J. Biotechnol.*, 9,15, (2010) 2267.
16. Guenther E., *VIII.Robert. E D. Von Nostrand comp.,Inc. New York* (1961).
17. Adams R.P., *Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry. 4th Edn., Allured Publishing Corp., Carol Stream, Illinois. , 1995*
18. Kovats E., *Helvetica Chimica Acta*, 41, (1958) 1915.
19. Piper C. S., *Inter Sci. Publishers Inc. New. York*. (1950),
20. Snedecor W. G., Cochran G. W., *Statistical Methods 6th Ed., The Iowa State Univ. Press, Iowa ,USA* (1967)
21. Rice G., editor-in-chief *American Horticultural Society Encyclopedia of Perennials. New York, NY: DK Publishing, Inc. .(2006).*

(2017) ; <http://www.jmaterenvironsci.com>