Effect of the nitrogen fertilizer forms and time of their application on the yield of herb and essential oil of *Ocimum americanum* L.

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S u m m a r y

The effect of various forms of nitrogen fertilizer and dates of application on *Ocimum amercianum* yield and essential oil (ton/feddan, 1 feddan = 4200 m^2) was studied in two successive seasons. In both seasons the total yields of fresh and dry herb after the application of ammonium sulfate surpassed that of ammonium nitrate and urea. Dividing the full amount of fertilizer into two equal portions gave the highest yields in most cuts.
Nitrogen fertilizers affected the percentage of essential oil in most cuts. The highest oil percentage was obtained after the application of ammonium nitrate and ammonium sulfate in both cuts in the first season, respectively, and urea in the second season. Early addition of the full amount of fertilizer was more effective in increasing oil percentage as compared to other two dates of application. Ammonium sulfate in the first season and urea in the second one produced the highest total oil yields either per plant or per feddan. One application of the full amount of ammonium nitrate at the early date produced the highest total percentage of monoterpenic compounds as compared to ammonium sulfate and urea. One application the full amount of nitrogen fertilizer at the early date was superior to other treatments. The opposite was true for sesquiterpenes. Oxygenated compounds showed the same trend of monoterpenic compounds.
Thus, the application of ammonium sulfate at the rate of 60 kg N/fed/season at two equal portions in each cut could be recommended for maximizing herb, leaves and oil yields of *Ocimum amercianum*, L. var. *pilosum* plants.

Key words: Ocimum amercianum L.*, essential oil, GLC, nitrogen sources, date of application*

INTRODUCTION

The *Ocimum* genus, *Labiatae* (*Lamiaceae*) family contains from 50 to 150 species of herbs and shrubs from tropical regions of Asia, Africa as well as Central and South America. It has been widely used as a medicinal and aromatic plant in many countries, i.e. Egypt, India, Greece, Italy, Morocco and other. *Ocimum americanum* L. (syn. *O. canum*), hoary basil or mosquito plant has three distinct chemotypes: floral-lemony, camphoraceous and spicy. The seed of *Ocimum americanum* L. was introduced by Cornell University through a scientific exchange and cultivated in Egypt for propagation and adaptation for the environmental conditions of this country. After two years of cultivation it was found that its essential oil is rich in eugenol and methyl chavicol [1]. The major inorganic forms of N absorbed by plants are NO_3^- and NH_4^+. Both forms of N can be presented naturally in the soil solution. Ammonium is the most often applied form of N fertilizers. It is rapidly nitrified to NO_3^- under most soil conditions. Ammonium nitrification relies mainly on soil texture and its pH, since soils of low pH and/or bad aeration arrested the rate of nitrification [2, 3]. It can be stated that N form (NO_3^- or NH_4^+) did not alter the percentage of monoterpenes and aromatic polypropanoides in *O. basilicum* essential oil, but NH_4^+–N increased the total sesquiterpene percentage. Sinha and Gulati [4] detected the presence of-α pinene, camphene, β-pinene, myrcene, p-cymene, 1,8-cineole, terpinolene, linalool, camphor, citronellal, iso-borneol, borneol, methyl chavicol, nerol, geraniol, thymol, eugenol, methyl eugenol, iso-eugenol, caryophyllene, humulene, acetoeugenol and V-cadinene in the essential oil of *O. americanum*. Also Upadhyay et al. [5] mentioned that the volatile oil of *O. americanum* (camphor-rich chemotype) afforded 9 monoterpene hydrocarbons, 4 oxygenated monoterpenes, 12 sesquiterpene hydrocarbons, 7 sesquiterpene alcohols and a few unidentified trace compounds. Four new alcohols of the uncommon copane series were also isolated and their structures elucidated by spectroscopic methods and chemical transformations. Gulati and Sinha [6] found that camphor, linalool and citral are three chemotypes of *O. canum* (*O. americanum*). Weaver et al. [7] mentioned that linalool was present at 8.6±0.9 mg/g in dried leaves of *O. americanum*. Gupta [8] indicated that the main component of the essential oil of *O. canum (O. americanum,* linalool-type) is linalool which reached its maximum concentration at 50% seed set stage. Shahi and Gupta [9] performed phenological studies on a linalool- rich strain of *O. canum* (*O. americanum*). It was found that linalool comprised 76% of the essential oil. Under the Egyptian conditions Omer et al. [1] cultivated *O. americanum* var. *pilosum* and studied the essential oil of the leaves with gas chromatography mass spectrometry (GC-MS). They found that eugenol (22.07%) was identified as the main compound followed by farnesene (14.79%), then 1,8-cineole (12.27%). Sesquiterpenes comprised 33.62% of the total oil content. Among sesquiterpenes, farnesene (14.79%) was found to be the main. They concluded that the oil of *O. americanum* cultivated in Egypt is eugenol-rich and postulated that it is due to a dual biosynthetic pathway.

This work aimed to study the effect of three nitrogen sources: ammonium nitrate (AN), ammonium sulfate (AS), and urea (U) and/or dates of application: early (E), early-late (EL), and late (L) on the yield and essential oil of *Ocimum americanum* L.

MATERIAL AND METHODS

This work was carried out in two successive seasons (2000 and 2001) at the Experimental Farm of Cultivation and Production of Medicinal and Aromatic Plants Department, National Research Centre at Boulak El-Dakrour, Giza, Egypt.

Samples from the soil were taken before cultivation and were subjected for physical and chemical analysis according to Jackson [10].The results indicated that the soil was clay-loam in texture.

In each season, the soil was mechanically ploughed and planked twice. During the soil preparation for cultivation a mixture of calcium super-phosphate (15.5% P_2O_5) as a source of phosphorus and potassium sulfate (48% K_2O) as a source of potassium was added at the rate of 200 and 100 kg/fed, respectively, and was manually mixed with the soil.

American basil seeds (*O. americanum* var. *pilosum*) were obtained from Cornell University and propagated in Egypt. The resulted seeds were sown on 1 March 2000 and 2001 in the nursery. They germinated after about three weeks. On 15 April 2000 and 2001 the uniform healthy basil seedlings (10 cm of length) were transplanted into the field on rows with spacing of 60 cm and 30 cm between hills. Surface irrigation with Nile water was used for irrigating plants as necessary. Weed control was carried out manually in the whole growing season.

The used nitrogen sources were ammonium nitrate (AN, 33.5% N), ammonium sulfate, (AS, 20.5% N) and urea (U, 46% N). Each source was applied at the rate of 60 kg N/fed. Half the amount of each fertilizer was added in the first cut and the other in the second cut. There were three dates of application in each cut. At the early date treatment (E) the full amount of nitrogen was applied once after one month of transplanting. At the early-late treatment (EL) the full amount of nitrogen was divided into two equal portions: one of them was applied after one month of transplanting and the other after two months of transplanting. The late date of application (L) involved the simultaneous addition of full amount of nitrogen after two months of transplanting. In the second cut the early addition was after two weeks of cutting and the late addition one month after the first one.

The used experimental design was a split plot design, where the treatments were distributed randomly within plots with four replicates of each treatment. The nine treatments were the combinations of three sources of nitrogen and three dates of application. Sources of nitrogen were situated in main plots and dates of application in the subplots. The area of every main plot was 15 m^2, while it was 5 m^2 for each sub-plot.

The plants were harvested two times in each growing season and then the plants were left for seed setting. The first cut was done on 15 July during the flowering stage, the second one on 1 October by cutting the vegetative parts of the plants 10 cm above the soil surface. Herb fresh and dry yield (ton/fed.) and leaves fresh and dry yield (ton/fed.) were determined for each replicate. Data of the present study were statistically analyzed according to Cochran and Cox [11].

The extraction of the essential oil was carried out according to Guenther [12] on the basis of the herb fresh weight at the laboratory of Cultivation and Production of Medicinal and Aromatic Plants, National Research Centre (NRC), Dokki, Cairo, Egypt. The resulted oil was dehydrated over anhydrous sodium sulfate and stored in glass vials at freezer in the absence of light till used for gas liquid chromatographic (GLC) analysis.

The GLC analysis of the oil samples was carried out in the second season using Hewlett Packard gas chromatograph apparatus at the Central Laboratory of NRC with the following specifications: instrument: Hewlett Packard 6890 series, column: HP (Carbwax 20M, 25 m length x 0.32 mm I.D), film thickness: 0.3 mm, sample size:1 µl, oven temperature: 60–190ºC, program: 60ºC/2 min, 8ºC/ min, 190ºC/25 min, injection port temperature :240ºC, carrier gas: nitrogen, detector temperature (FID): 280ºC, flow rate: N2 30 ml/min., H_2 30 ml/min., air 300 ml/min. Main compounds of the essential oil were identified by matching their retention times with those of the authentic samples injected under the same conditions. The relative percentage of each compound was calculated from the peak area corresponding to each compound.

RESULTS AND DISCUSSION

The fresh yield (ton/fed.) responded significantly to nitrogen fertilizer forms in both seasons with exception for the first cut of the second season (tab. 1). Ammonium sulfate was superior to the other two nitrogen forms in biomass production giving the highest mean values. The differences in the herb fresh yield due to ammonium sulfate and ammonium nitrate were significant in both seasons, while the differences due to ammonium sulfate and urea were significant only in the first season. Although the effect of nitrogen forms was insignificant in the first cut of the second season, the differences due to ammonium nitrate and urea were significant in both seasons, in which the superiority was for ammonium nitrate in the first season and for urea in the second season. In all cases, the total yield of both cuts of each season showed the same trend in the second cut. The total yield of the fresh herb in the second season was higher than that of the first season by 28.3%. The second cut produced fresher yield than those of the first cut by 11.66 and 19.9% in the first and second seasons, respectively. The mean values of date of application and portions of fertilization showed significant differences in the yield of dry herb in both cuts of both seasons in addition to total yield. Except from the first cut of the first season, splitting the full amount of each fertilizer into two equal portions produced higher yields of fresh herb as compared to single fertilization either at early or at a late date. The effect of interaction between nitrogen sources, date and number of application was significant in both seasons, except from the first cut of the second season. Splitting the full amount of ammonium sulfate into two equal portions resulted in the highest yields of the fresh herb in the second cuts of the first and second seasons, respectively, while the smallest yields resulted from single application of the full amount of ammonium nitrate at the late date in the first cuts of both seasons, respectively. It is clear that, maintaining low but adequate concentration in the soluble N pool, applying nitrogen fertilizer in times of increased uptake capacity or both would have been a possible means of improving nitrogen uptake efficiency and subsequently increased the metabolic capacity of plants which resulted in higher biomass production. The present study is in accordance with other findings reported by Mousa [13] on *Ocimum canum*. They all showed that ammonium sulfate increased the fresh yield of herb.

Table 1.

Herb fresh yeld (ton/fed) and herb dry yield (ton/fed) of *Ocimum americanum* L. as affected by different nitrogen sources and date of application in two seasons.

treatments	herb fresh yield (ton/fed.)						herb dry yield (ton/fed.)					
	1st season (2000)			2nd season (2001)			1st season (2000)			2nd season (2001)		
	1st cut	2nd cut	total	1st cut	2nd cut	total	1st cut	2nd cut	total	1st cut	2nd cut	total
E	13.18	13.84	27.02	15.56	18.84	34.39	2.593	2.745	5.338	3.112	3.903	7.015
AN EL	12.92	14.78	27.70	17.11	20.44	37.55	2.522	3.018	5.540	3.421	4.085	7.506
L	10.32	13.48	23.80	13.78	15.77	29.55	2.454	2.716	5.170	2.849	3.440	6.289
mean	12.14	14.03	26.17	15.48	18.34	33.82	2.523	2.826	5.349	3.127	3.809	6.936
E	12.67	14.69	29.36	16.46	20.14	36.60	2.593	2.943	5.886	3.340	4.065	7.405
AS EL	13.86	16.21	30.07	17.26	21.65	38.91	2.522	3.219	6.062	3.452	4.250	7.702
L	11.94	13.74	25.68	15.95	18.02	33.97	2.454	2.787	5.294	3.236	3.723	6.959
mean	13.49	14.88	28.37	16.55	19.93	36.48	2.764	2.983	5.747	3.342	4.012	7.353
E	11.67	12.63	24.30	16.19	19.24	35.43	2.636	2.593	5.229	3.301	3.856	7.157
U EL	11.67	13.63	25.30	16.97	20.32	37.29	2.584	2.716	5.300	3.369	4.166	7.535
L	10.89	11.10	21.99	14.74	18.31	33.05	2.304	2.456	4.76	3.067	3.695	6.762
mean	11.41	12.45	23.86	15.96	19.29	35.25	2.508	2.588	5.096	3.245	3.905	7.150
total mean	12.34	13.78	26.13	15.99	19.18	35.18	2.598	2.799	5.397	3.238	3.908	7.146
E	13.17	13.72		16.07	19.40		2.724	2.760		3.251	3.941	
mean EL	12.81	14.87		17.11	20.80		2.649	2.984		3.414	4.167	
L	11.05	12.77		14.82	17.36		2.421	2.653		3.050	3.619	
LSD at 5%												
sources	0.456	0.353		NS	0.728		0.083	0.063		0.161	0.104	
dates	0.619	0.379		0.632	0.605		0.076	0.081		0.124	0.071	
interaction	1.072	0.657		NS	1.048		0.132	NS		NS	NS	

Data presented in table 1 reveal that ammonium sulfate significantly increased herb dry yield compared to other two nitrogen sources with exception for the first cut in the second season in which the differences between ammonium sulfate and urea were insignificant. The maximum mean values of total yields were obtained due to ammonium sulfate. Meanwhile, the minimum mean values of total yields were obtained due to urea and ammonium nitrate in the first and second season, respectively. Application of ammonium nitrate and urea showed no significant differences in most cases.

Taking into consideration the effect of application dates on herb dry yield, results showed a variable trend upon the two cuts of the first season. Applying all the amount of nitrogen at the same time at the early date produced the highest mean value, while dividing the full amount of nitrogen into two equal portions gave the highest mean value in the second cut. On the other hand, the effect of interaction between nitrogen sources and dates of application was insignificant in both seasons, with exception for the first cut of the first season. The maximum value was obtained due to ammonium sulfate added twice in two equal doses in the second cut of the second season. Similar results were recorded by Mousa [13] on *Ocimum canum* as they all indicated that ammonium sulfate increased herb dry yield.

From the above mentioned results it may be concluded that splitting the full amount of ammonium sulfate into two equal portions in each cut is the best recommended treatment for maximizing fresh and dry yield of herb in *O. americanum* var. *pilosum* plants. On the other hand, the early application of ammonium sulfate enhanced leaves production. Similar results were obtained by Omer et al. [1] and Youssef et al. [14] on *Ocimum basilicum* plant.

Sources of nitrogen significantly affected the essential oil percentage in the fresh herb of basil in the first season and the first cut of the second season (tab. 2). In the first cut of the first season ammonium nitrate significantly increased oil percentage giving the highest value followed by urea and then by ammonium sulfate which produced the least oil percentage. In the second cut an opposite trend was observed, as ammonium sulfate gave the highest oil percentage followed by urea and then by ammonium nitrate which gave the least oil percentage. Application of ammonium nitrate and urea showed no significant differences in both cuts. In the second season application of urea significantly increased the oil percentage as compared to ammonium nitrate and ammonium sulfate used in the first cut. The difference between ammonium nitrate and ammonium sulfate was insignificant. Oil percentage revealed no significant response to all nitrogen sources in the second cut. Although no significant differences were noticed among the date and number of portions in the first cut of the first season, adding the full amount of nitrogen at the same time at the late date led to higher percentage than other two treatments. In the second cut, applying the full amount of nitrogen at the same time at the early date seemed to be more effective in increasing oil percentage as compared to other two treatments. On the other hand, adding the full amount of nitrogen at the late date resulted in the least percentage in the second cut of the first season.

Table 2.

Effect of different nitrogen sources and date of application on *Ocimum americanum* L. essential oil percentage in two seasons

treatments	1st season (2000)		2nd season (2001)	
	1st cut	2nd cut	1st cut	2nd cut
E	0.238	0.183	0.227	0.190
AN EL	0.227	0.170	0.210	0.195
L	0.245	0.173	0.205	0.230
mean	0.237	0.175	0.214	0.205
E	0.232	0.203	0.222	0.227
AS EL	0.218	0.205	0.202	0.218
L	0.200	0.197	0.193	0.200
mean	0.217	0.202	0.206	0.215
E	0.215	0.197	0.258	0.230
U EL	0.238	0.203	0.278	0.225
L	0.245	0.175	0.202	0.170
mean	0.232	0.192	0.246	0.208
total mean	0.228	0.189	0.222	0.209
E	0.228	0.194	0.235	0.215
mean EL	0.227	0.192	0.230	0.212
L	0.230	0.181	0.200	0.200
LSD at 5%				
sources	0.012	0.018	0.012	NS
dates	NS	0.008	0.016	0.012
interaction	0.024	NS	0.028	0.022

In the second season, the results were rather similar in both cuts. Adding the full amount of nitrogen at the early date significantly increased the oil percentage as compared to adding full amount at the late date. Whereas, these increase failed to reach the level of significance compared to splitting the full amount of nitrogen into two portions. Therefore, the highest oil percentages were obtained from the application of the full amount of nitrogen at the early date.

The interaction between nitrogen sources and dates of application was significant in both seasons except from the second cut of the first season. The highest value was obtained from application of urea added twice in two equal portions in the first cut of the second season. Mousa [13] declared that ammonium nitrate gave the highest oil percentage in *Ocimum canum*.

The relation between dates of application of nitrogen sources and oil yield (ml/plant) is shown in table 3. Nitrogen sources had a beneficial and significant effect on oil yield in both seasons. In the first season, ammonium sulfate had the most distinct role in producing more oil yields followed by ammonium nitrate, while urea produced the least oil yields. Therefore, the highest mean value of total seasonal oil yield was obtained due to ammonium sulfate as an evident of its superiority in both cuts.

In the second season, data showed a variable trend during the two cuts, while the highest mean oil yield was observed from urea in the first cut, it was observed from ammonium sulfate in the second cut. On the contrary, in the first season the total oil yield after the use of urea was superior to those of ammonium nitrate and ammonium sulfate in the second season. The increments reached 13.42 and 3.73%, respectively.

Regarding the effect of dates of application on oil yield (ml/plant) it was found that dividing the full amount of nitrogen into two equal portions was more effective in yielding more oil in both seasons, except from the first cut of the first season in which simultaneous application of the full amount of nitrogen resulted in the highest mean value. Although, the differences between these two treatments were insignificant in the first cut of both seasons. In all cases, one adding of the entire amount at the late date markedly reduced the mean value of oil yield as compared to the other two dates of application. The interaction effect between nitrogen sources and dates of application was significant in both cuts of both seasons. The highest value was obtained either by application of ammonium sulfate or urea in two equal doses in the first or second cut of the second season. On the other hand, the minimum oil content was produced from plants received the full amount of urea once at the late date in the second cut of the first season.

The total mean of the two seasons indicated that mean of the second cut was higher than that of the first cut. In general, differences in oil yield among the cuts during both growing seasons can probably be attributed to differences in herbage yield due to differences in photosynthesis, temperature and moisture conditions. Light levels were especially high during the growth period of the second cut (July to October) which contained the higher oil yield. Pitarevic et al. [15] observed an increase in oil production in *Salvia officinalis* under drought conditions. Omer et al. [16] reported that the harvest of marjoram in October gave the highest oil percentage and yield. The positive effect of ammonium sulfate on increasing oil content was confirmed by the findings of Mousa [13] on *Ocimum canum.* On the other hand, Alder et al. [3] stated that ammonium nitrogen form decreased the oil yield of *Ocimum basilicum*.

The response of oil yield (l/fed.) to nitrogen sources and dates of application is shown in table 3. Results indicate that the response was rather similar to that of oil yield (ml/plant), in which the oil yield significantly responded to sources of nitrogen, date of application and their interaction. In the first season, application of ammonium sulfate increased oil yield per feddan comparing to ammonium nitrate and urea. The differences were significant, except from the difference between ammonium nitrate and ammonium sulfate in the first cut. The highest mean values were obtained with ammonium sulfate followed by ammonium nitrate and then by urea which gave the least mean values. Consequently, the total seasonal oil yield of ammonium sulfate was superior to those of ammonium nitrate and urea by 11.58 and 17.68%, respectively.

Table 3.

Effect of different nitrogen sources and date of applications on the essential oil content (ml/plant) and essential oil yield (l/fed) of *Ocimum americanum* L. in two seasons

treatments		essential oil content (ml/plant)						essential oil yield (l/fed.)					
		1st season 2000)			2nd season (2001)			1st season (2000)			2nd season (2001)		
		1st cut	2nd cut	Total	1st cut	2nd cut	Total	1st cut	2nd cut	Total	1st cut	2nd cut	Total
AN	E	1.659	1.337	2.996	1.865	1.887	3.752	31.35	25.32	56.67	35.32	35.78	71.10
	EL	1.551	1.325	2.876	1.900	2.105	4.005	29.32	25.13	54.45	35.93	39.86	75.79
	L	1.337	1.230	2.567	1.490	1.917	3.407	25.27	23.32	48.59	28.23	36.28	64.51
	mean	1.515	1.297	2.813	1.751	1.969	3.721	28.64	24.59	53.23	33.16	37.30	70.46
AS	E	1.800	1.575	3.375	1.930	2.415	4.345	34.01	29.81	63.82	36.54	45.72	82.26
	EL	1.599	1.755	3.354	1.840	2.492	4.332	30.22	33.22	63.44	34.86	47.19	82.05
	L	1.264	1.425	2.689	1.625	1.905	3.530	23.89	27.07	50.96	32.21	36.05	68.26
	mean	1.554	1.585	3.139	1.798	2.270	4.069	29.37	30.03	59.41	34.53	42.98	77.52
U	E	1.327	1.312	2.639	2.207	2.337	4.544	25.08	24.87	49.95	41.77	44.28	86.05
	EL	1.469	1.462	2.931	2.492	2.417	4.909	27.75	27.66	55.41	47.18	45.72	92.90
	L	1.411	1.025	2.436	1.570	1.640	3.210	26.66	19.42	46.08	29.77	31.12	60.89
	mean	1.402	1.266	2.668	2.089	2.131	4.220	26.49	23.98	50.48	39.57	40.37	79.94
total mean		1.490	1.382	2.873	1.879	2.123	4.003	28.16	26.20	54.37	35.75	40.21	75.97
mean	E	1.595	1.408		2.000	2.213		30.14	26.66		37.87	41.92	
	EL	1.539	1.514		2.077	2.338		29.09	28.67		39.32	44.25	
	L	1.337	1.226		1.561	1.820		25.27	23.27		30.07	34.48	
LSD at 5%:													
	sources	0.044	0.031		0.148	0.083		1.012	0.663		2.820	1.566	
	dates	0.071	0.027		0.076	0.066		1.379	0.683		1.804	1.331	
	interaction	0.124	0.046		0.132	0.115		2.389	1.183		3.124	2.306	

In the second season, the highest mean values of oil yields were obtained as a result of application of urea and ammonium sulfate, respectively. The response of total seasonal oil yield (l/fed) was rather similar to the response of the first cut. It means that the highest mean value of total seasonal oil yield (79.94 l/fed.) was obtained from plants fertilized with urea, while the least mean value of total oil yield (70.46 l/fed.) was obtained from plants fertilized with ammonium nitrate. The differences between ammonium nitrate and ammonium sulfate were insignificant in the first cut only.

Regarding the effect of dates and portions of application on oil yield per feddan, results showed a variable trend during the two cuts of the first season: adding the full amount of nitrogen at the early date insignificantly increased the mean value of oil yield as compared to dividing the full amount into two equal portions in the first cut. In the second cut the opposite was true, where dividing the full amount of nitrogen significantly increased the mean value of oil yield compared to application of the all amount either at the early or at the late date.

Relatively similar trend was obtained in both cuts of the second season. The highest mean values of oil yields were obtained as a result of dividing the all amount of nitrogen into two equal portions. Whereas, delaying the application time significantly reduced the mean values of oil yield in all cuts of both seasons.

Oil yield per feddan showed a significant response to the interaction effect between nitrogen sources and dates of application in both cuts of both seasons.

The highest value was obtained with application of ammonium sulfate in two equal portions in the second cut of the second season. The minimum value was produced from adding the full amount of urea in a one moment at the late date in the second cut of the first season. Several investigators, such as Youssef et al. [14], studied this point of research on *Ocimum basilicum* and stated that ammonium nitrate gave the highest oil yield.

The main constituents of the essential oil of the second cut of the second season as affected by different nitrogen fertilizer forms and number and date of application are shown in table 4. In all treatments, eugenol was identified as a major compound and ranged from 33.68% for ammonium nitrate added once at the late date to 39.42% for ammonium nitrate added at two equal splits. For ammonium nitrate and ammonium sulfate, splitting the full amount into two equal portions was superior than one adding of the full amount either at late or at early time. For urea one adding the full amount at the early date increased eugenol content. The differences in eugenol content as a result of different nitrogen sources were not considerable and did not exceed 4.0%. Methyl chavicol content was also altered by different nitrogen sources. Although the differences were not considerable, urea showed the highest mean value of methyl chavicol content (15.35%) as comapred to the other two nitrogen sources which exceeded by 5.06 and 13.03% more than ammonium nitrate and ammonium sulfate, respectively. There was no considerable effect of date and number of application on methyl chavicol content.

Table 4

The relative percentage of the main constituents detected with GLC in the oil of *Ocimum americanum,* L. plants as affected by different nitrogen sources and date of applications (the 2nd cut of the 2nd season)

Components	RRt	AN			AS			U		
		E	EL	L	E	EL	L	E	EL	L
α-pinene	0.110	0.399	1.201	trace	0.453	0.412	0.297	trace	trace	0376
β-pinene	0.128	1.358	trace	1.034	1.104	1.443	0.706	trace	trace	1.029
myrcene	0.150	1.135	trace	1.001	1.379	1.217	1.025	0.491	0.401	1.151
α-terpinene	0.178	0.298	trace	0.284	0.345	trace	0.306	trace	trace	0.304
limonene	0.216	7.820	8.190	7.905	8.494	8.343	7.939	7.797	7.082	8.601
1,8-cineole	0.244	7.886	7.133	6.788	6.448	6.469	5.561	5.674	4.583	6.605
camphor	0.528	1.289	1.390	1.456	1.282	1.212	1.324	1.516	1.614	1.347
linalool	0.539	1.656	1.012	1.823	1.620	1.377	1.727	1.702	1.660	1.549
linalyl acetate	0.579	0.584	trace	0.638	0.533	0.520	0.560	0.622	0.650	0.556
farnesene	0.606	8.258	9.804	9.536	9.235	8.890	9.408	10.17	10.93	9.280
β-bisabolene	0.637	3.988	3.431	3.869	3.508	3.363	3.957	3.101	3.619	4.017
methyl chavicol	0.658	14.13	14.05	15.65	13.45	12.84	14.45	15.84	16.62	13.61
terpineol	0.697	12.29	12.05	12.58	10.94	10.60	11.23	12.37	13.13	11.76
methyl eugenol	0.881	0.247	trace	0.356	0.277	0.363	0.309	trace	trace	0.259
eugenol	1.000	36.36	39.42	33.68	38.43	39.57	36.45	38.00	36.64	37.15
iso-eugenol	1.026	0.722	0.821	0.818	0.666	0.806	0.739	0.822	0.917	0.690
farnesol	1.137	0.766	0.760	0.520	0.678	0.801	0.592	0.688	0.647	0.633

Terpineol occupied the third position in the range of major compounds of the essential oil, after eugenol and methyl chavicol, with total mean value of 11.88%. Ammonium sulfate resulted in the minimum mean value of terpineol (10.92%), while the mean values of ammonium nitrate and urea were close one to another (12.4%). Time and date of nitrogen application showed no considerable differences in terpineol content.

The relative percentage of farnesene in the essential oil ranged from 8.258% for ammonium nitrate applied at the early date to 10.17% for urea applied at the early date. It was clear that urea surpassed ammonium nitrate and ammonium sulfate in increasing farnesene content giving the highest mean value (10.129%). Splitting the full amount of the fertilizer into two equal doses recorded the highest mean value (9.88%) followed by late application of the full amount of the fertilizer (9.408%).

Plants fertilized with ammonium nitrate were more effective in increasing 1,8-cineole content, recording the highest mean value (7.269%) in comparison to ammonium sulfate (6.159%) and urea (5.620%). Single application of the full amount of nitrogen fertilizer at the early date gave the highest mean value (6.669%) followed by the application at the late date (6.318%).

Plants fertilized with different nitrogen sources showed no considerable differences in limonene content. Ammonium sulfate gave a slight increase in the mean value of limonene content compared to ammonium nitrate and urea. The dates of application also revealed no considerable effect on limonene content. One application of the full amount of fertilizer at the late date led to a slight increase.

The total percentage of monoterpenic compounds accounted 82.62% with one addition of the full amount of ammonium sulfate at the late date and 86.18% with one application of the full amount of ammonium nitrate at the early date. Ammonium nitrate produced more monoterpenic compounds than ammoniun sulfate or urea. One appliance of the full amount of nitrogen at the early date was superior to the other treatments in production of monoterpenoides. The opposite was true for sesquiterpenes. Oxygenated compounds showed the same trend of monoterpenic compounds.

We may recommend the fertilization of *Ocimum americanum* L. var. *pilosum* with 60 kg N/fed. (equal to 292 kg of ammonium sulfate/fed./season) for maximizing the production of its herb and leaves and the essential oil. It could be recommended also to add half the amount of the fertilizer in the first cut at two equal portions; the first portion added after one month of transplanting, the other after two months of transplanting. The remainder can be added in the second cut at two equal portions; the first portion after two weeks of cutting and the other after one month and half from cutting.

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WPŁYW RODZAJÓW NAWOZÓW AZOTOWYCH I CZASU ICH ZASTOSOWANIA NA PLON ZIELA I OLEJEK ETERYCZNY BAZYLII (*OCIMUM AMERICANUM* L.)

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S t r e s z c z e n i e

W dwóch następujących po sobie okresach wegetacji badano wpływ różnych rodzajów nawozów azotowych i czasu ich zastosowania na plon ziela i olejku eterycznego bazylii (*Ocimum americanum* L.) mierzony w tonach z feddana (1 feddan = 4200 m^2 = 1,038 ara).

W obu okresach wegetacji całkowity plon ziela i suchej masy po zastosowaniu siarczanu amonu przewyższał ten uzyskany po zastosowaniu azotanu amonowego i mocznika. Dzielenie całej porcji nawozu na dwie równe części spowodowało najwyższe plony podczas większości zbiorów.

Rodzaje zastosowanych nawozów azotowych wpływały na procentową zawartość olejków eterycznych w zielu podczas większości zbiorów. Najwyższą zawartość olejków eterycznych zanotowano po zastosowaniu odpowiednio siarczanu amonu i azotanu amonowego w obu zbiorach w pierwszym okresie wegetacji i mocznika w drugim. Wczesne zastosowanie całej porcji nawozu bardziej podnosiło zawartość olejku eterycznego w porównaniu z dwoma innymi terminami ich zastosowania. Zastosowanie siarczanu amonu w pierwszym okresie wegetacji i mocznika w drugim dało największy całkowity plon olejku zarówno w pojedynczej roślinie, jak i z feddana. Jednorazowe zastosowanie całej porcji siarczanu amonu we wczesnym okresie dało największy możliwy plon składników monoterpenów w porównaniu z siarczanem amonu i mocznikiem. Jednokrotne zastosowanie całej porcji nawozu azotowego we wczesnym okresie przewyższało inne rodzaje nawożenia. Odwrotnie było w wypadku seskwiterpenów. Zastosowanie składników utlenionych wykazało tendencję podobną do tej obserwowanej w wypadku monoterpenów.

Można więc polecić zastosowanie siarczanu amonu w ilości 60 kg/fed./sezon w dwóch równych porcjach dla każdego zbioru w celu zwiększenia plonu ziela, liści i olejku eterycznego bazylii.

Słowa kluczowe: Ocimum americanum L., *olejek eteryczny, GLC, źródła azotu, terminy nawożenia*