Effect of sowing date and application of foliar fertilizers on the yield and chemical composition of rue (*Ruta graveolens* L.) herb

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S u m m a r y

The present investigation was conducted during two successive seasons of 2004/2005 and 2005/2006 in order to study the effect of two different sowing times and two macro- and microfoliar fertilizers on yield and chemical composition of *Ruta graveolens* plants. Delay in sowing from 20 October to 20 November resulted in significant promotion for herb dry weight (g/m^2) as well as percentage and yield of essential oil of herb. In contrast, the accumulation of rutin and coumarin contents was higher at the first sowing time than the second one. The main constituent of essential oil in herb was identified as 2-undecanone. It represented 60.74% of identified herb essential oil at first sowing date and decreased to 55.88% at second date.
Both foliar fertilizers, Crestalon and Leaf drip, promoted herb dry weight. Leaf drip was more favorable to improve these criteria than Crestalon. In general, the highest content of essential oil, rutin and coumarin were recorded with application of 1000 or 2000 ppm of Leaf drip. The maximum yield of herb and essential oil resulted in plants of the second date of sowing and fertilized with Leaf drip at 2000 ppm.

Key words: rue, foliar fertilizers, sowing date, essential oil, rutin, coumarin

INTODUCTION

Ruta graveolens is an herbaceous plant originally native to the Mediterranean region and cultivated in many parts of the world. Rue's odor is considered to be repulsive. There are two main species used in traditional medicine, *Ruta chale-*

pensis and *R. graveolenus* [1]. Rue's application in herbal therapy was to promote menstruation [2], relieve symptoms of hangover [3] as contraceptive [4]. It is also applied externally as poultice against rheumatic pain [5]. Rue's flavonoid content possesses antibacterial activity [6] as well as cytoxic effects *in vitro* [7]. Moreover, Chiu and Fung [8] indicated that rue plants contained cardiovascular active substances that had a direct effect on the cardiovascular system.

Foliar nutrition is widely used in order to correct specific nutrient deficiency or to prove nutrient, what is preferable especially in newly reclaimed soil. Plants response to foliar nutrition varies according to several factors such as plant species and environmental conditions. Several researches reported the beneficial effect of foliar fertilization on growth and yield of different medicinal and aromatic plants [9-12].

The effect of sowing date on growth, yield and active ingredients of medicinal and aromatic plants was studied by many investigators [13-15].

This paper aimed to study the effect of sowing date and foliar nutrition on yield and active constituents of *Ruta graveolens*.

MATERIAL AND METHODS

Two field experiments were conducted at the National Research Centre Farm in Shalakan, Kalubia Governorate, Egypt in two successive seasons of 2004/2005 and 2005/2006 in order to study the effect of different sowing dates on herb and active substances of *Ruta graveolens* plants treated with two foliar fertilizers (Crestalon and Leaf drip).

Materials

Seeds of rue plants were obtained from the Experimental Farm of Pharmaceutical Factory at Giza and planted during two successive seasons at two sowing dates (20 October and 20 November). The seeds were planted in plots of 2 x 2 m^2, ^{e}ach plot of 3 rows with 40 cm distance between plants. Foliar application of Crestalon (Manufactured by Agrotech, Co. Phythotreptid Co., Modern Agriculture) and Leaf drip (Frarinysex, France) were carried out twice: the first one was applied after two months of cultivation and the second one was one month later. The chemical compositions of the two foliar fertilizers were presented in table 1, while the physical and chemical properties of soil are shown in table 2, using the method described by Black [16].

Four doses of two foliar fertilizers were applied: 0, 1000, 2000 and 3000 ppm of each. Topal was added (1 ml/L) as a wetting agent. The control treatment (0 ppm) was sprayed with distilled water at the same time. All other agricultural practices were done as needed. The design of the experiment was split plot, with 3 replicates. The sowing dates were in the main plot and the two foliar fertilizers were in the sub-plot.

Table 1.

Chemical composition of the two foliar fertilizers of Crestalon and Leaf drip

fertilizer	total N	phosphate	Pot.	Mg	Br	Fe	Mo	EDTA, SO_3	Cu	Mn	Zn
Crestalon	19%	19%	19%	-	0.25%	–	1 gm/L	–	–	–	–
Leaf drip	19%	19%	19%	1%	70 ppm	100 ppm	15 ppm	1.7%	15 ppm	150 ppm	70 ppm

Table 2.

Some physical and chemical characters of experimental soil

soil characters	value	soil characters	value
sand (%)	48.8	E.C. (m. mohs/cm)	0.68
silt	28.0	N (%)	0.11
clay	24.2	P (mg/100 g)	2.08
texture class	sandy loam	K (mg/100 g)	22.4
organic matter (%)	2.07	Zn (ppm)	1.0
pH	8.08		

Sampling and chemical analysis

Samples of plants representing two sowing dates were collected on 26 May and 28 June for the first and second seasons, respectively and the dry weight of herb (g/m^2) was recorded.

Samples of fresh herb of each treatment were subjected separately for hydro-distilation for three hours in order to extract and determine the percentage of essential oil, according to Guenther [17]. The essential oil resulted from each treatment was dehydrated over anhydrous sodium sulfate and kept in refrigerator till GC-MS analysis.

The gas chromatography mass spectrometry analysis of the essential oil samples was carried out using Varian Gas Chromatography (Walnut Creek, California, USA) equipped with Finnegau mat SSQ 700 (Therma Inst., USA) mass spectrometer with the use of following conditions:

– column: 30m x 0.25 mm DB-5 capillary column with film thickness (J&W Scientific, USA). The column temperature was programmed from 50°C (constant for 3 min.), at a rate of 70°C/min to 250°C with 10 min. isothermal hold.
– The injector temperature was 220°C and the transition temperature was 250°C. The carrier gas was helium and the column head pressure was 70–100 KPa.
– The identification of the constituents was determined by comparing the spectrum with the other stored in Wiley Mass Spectral Library containing over 147,000 volatile compounds.

Rutin and coumarin (as coumarin and furanocoumarins) content of herb were determined as mentioned by Zummo et al. [18] and Harbone [19], respectively.

Statistical analysis

The values of estimated parameters in two seasons were averaged and the mean values were statistically analyzed according to Snedecor and Cochran [20]. The values of L.S.D. at the level of 5% were assessed whenever the calculated "F" values were significant at the same level.

RESULTS AND DISCUSSION

Herb dry weight (g/m^2)

Effect of sowing date

Herb dry weight was significantly increased for the second sowing date at 20 November as compared to the first sowing date (tab. 3). The increment in the herb dry weight resulted from sowing on 20 November reached 50.6% compared to sowing on 20 October.

Many researchers, such as Omidbaigi and Mastro [15], demonstrated a strong influence of sowing time on growth character on Buck wheat. He recorded that the higher dry herb was produced from July and August sowing time, respectively. Singh and Randhawa [21] stated that delay in sowing from 15 to 30 September increased the herbage yield significantly. Zayed et al. [14] found that various growth parameters and yield of borage were decreased as the sowing date was delayed.

Effect of foliar fertilizer

Data tabulated in table 3 illustrated that both foliar fertilizers caused a corresponding increase in herb dry weight. Application of Leaf drip was more effective for this character than Crestalon at various levels of appliance. Thus, the highest increment of dry weight was noticed with Leaf drip at a level of 2000 ppm which caused highest promotional effect over the control treatment reaching 53.2%.

The promotive effect of foliar nutrients on growth characters were stated by several researches applying various foliar fertilizers [10, 11, 22, 23].

Effect of the interaction of sowing date and foliar fertilizers

Data tabulated in table 3 emphasized that various Leaf drip levels at second sowing date produced heaviest dry weight of herb. The highest values for these characters were obtained from application of Leaf drip at 2000 ppm at 20 November sowing.

Table 3.

Effect of sowing date and application of foliar nutrients on herb yield and it's chemical constituents of *Ruta graveolens* plants (mean values of two seasons)

	Fertilizer treat [ppm]	herb dry wt. [g/m^2]	essential oil [%]	essential oil yield [ml/m^2]	rutin [%]	coumarin [%]
1st sowing date	control	101.4	0.063	0.102	1.8	0.022
	Crestalon 1000	123.0	0.088	0.201	2.2	0.022
	Crestalon 2000	107.4	0.076	0.144	2.2	0.024
	Crestalon 3000	111.6	0.077	0.162	1.9	0.025
	Leaf drip 1000	130.2	0.068	0.189	1.9	0.025
	Leaf drip 2000	186.6	0.072	0.292	2.0	0.025
	Leaf drip 3000	181.8	0.068	0.265	2.1	0.026
means for 1st sowing date		134.4	0.073	0.194	2.01	0.024
2nd sowing date	control	175.2	0.119	0.679	1.7	0.020
	Crestalon 1000	181.8	0.147	0.855	1.7	0.020
	Crestalon 2000	187.2	0.150	0.935	2.0	0.022
	Crestalon 3000	181.2	0.149	0.904	2.0	0.023
	Leaf drip 1000	229.2	0.139	1.037	1.9	0.024
	Leaf drip 2000	237.6	0.174	1.739	2.1	0.024
	Leaf drip 3000	217.8	0.164	1.125	2.2	0.023
means for 2nd sowing date		202.2	0.149	1.025	1.94	0.022
mean values of fertilizer treatments	control	138.6	0.091	0.466	1.75	0.021
	Crestalon 1000	152.4	0.118	0.477	1.85	0.021
	Crestalon 2000	147.6	0.113	0,459	2.10	0.023
	Crestalon 3000	148.8	0.113	0.380	1.95	0.024
	Leaf drip 1000	180.0	0.103	0.491	1.90	0.025
	Leaf drip 2000	212.4	0.123	0.682	2.05	0.025
	Leaf drip 3000	198.0	0.116	0.490	2.15	0.025
L.S.D. fertilizer		3.41	0.005	0.031	------	-------
L.S.D. sowing date		2.92	0.003	0.083	-------	-------
L.S.D. fertilizer x sowing date		4.65	0.007	0.099	-------	--------

Essential oil content in herb

Effect of sowing date

Data indicated that delaying sowing date from 20 October to 20 November resulted in the highest promotional effect on essential oil percentage (tab. 3). The yield of essential oil in herb was affected by sowing date significantly. The second sowing date gave the maximum yield of essential oil for herb and this increment reached 428.3%. D· Antuono et al. [24] stated that oil yield of *N. sativa* decreased with delayed sowing. Si and Walton [25] reported that later sowing date resulted in the oil concentration of canola and Indian mustard was reduced by 1.1 percentage points for every two-week-delay in sowing.

Effect of foliar fertilizer

Data presented in table 3 show that foliar application of both fertilizers increased herb essential oil (%). The maximum mean value of essential oil percentage was obtained as a result of Leaf drip application at 2000 ppm followed by Crestalon at 1000 ppm.

It can be concluded that the application of Leaf drip was more effective and favorable for producing the highest accumulation of essential oil. The promotion in essential oil percentage and yield by both foliar fertilizers Crestalon and Leaf drip may be due to the effect on enzymes activity on metabolism improvements. Similar results were reported by Refaat and Balbaa [26] on *Cymbopogon flesuosus* Stapf; Khalil et al. [9] on Sinapis alba and *Nigella sativa*;Wahba and Ezz El-Din [10] on *Chrysanthemum coronarium*.

Effect of interaction between sowing date and foliar fertilizers

Essential oil content (%) and yield (ml/m^2) in herb showed various pronounced increments as a result of the combined treatment consisting of sowing dates and foliar application of both fertilizers (table 3). The combination of 2nd sowing date and foliar application of Leaf drip at 2000 ppm gave the maximum mean value of essential oil content (0.174) and yield (1.739ml/m^2).

Moreover, it is clear that all treatments of the second sowing date and various foliar fertilizers produced more herb essential oil than at the first date of sowing.

Rutin and coumarin content

Effect of sowing date

Rutin and coumarin content in herb of rue plant was affected by sowing time (tab. 3). Sowing of rue on 20 October was favorable for rutin and coumarins production. The difference between two sowing times for two compounds reached 3.6% and 8.1% for rutin and coumarin, respectively.

Effect of foliar fertilizer

Data in table 3 show that foliar application of both fertilizers increased rutin content in herb of rue plants. The highest mean value of rutin was obtained with the application of Leaf drip at 3000 ppm which passed the control by 22.8%. It could be noticed that Leaf drip was superior in this respect and the lower doses of both foliar fertilizer resulted in a lower accumulation of rutin.

The data also show that various levels of Crestalon or Leaf dirp caused insignificant accumulation of coumarin. All Leaf drip levels, which produced similar mean values of coumarin percent were higher than those of Crestalon levels.

Effect of the interaction between sowing date and foliar fertilizer

Data presented in table 3 revealed that the interaction between foliar application of both fertilizers and sowing dates generally caused promotive effect on the accumulation of rutin and coumarin.

The trend of increment of both substances was inconstant. However, the maximum percentage of rutin content was recorded from sowing date of 20 October with foliar application of Crestalon at a level of 1000 or 2000 ppm. Similarly, sowing at first date (20th October) and treatment of Leaf drip at 3000 ppm produced a highest percentage of coumarin.

Essential oil constituents

Effect of sowing date and foliar nutrient on herb essential oil constituents

The main constituents of essential oil of *Ruta graveolens* herbs as affected by different sowing date and application of foliar fertilizers (only the best treatments produced highest essential oil percentage) are shown in table 4. The identified compounds ranged from 98.59% to 99.88% of total compound. The hydrocarbonic compounds composed about 10.23% to 12.135%, while the oxygenated ones were 86.53% to 88.36% of the identified compounds.

Table 4.

Effect of sowing date and application of foliar nutrients on herb essential oil constituents of *Ruta graveolens* L.

compound	herb (first date)			herb (second date)			mean
	control	Crestalon 1000 ppm	Leaf drip 2000 ppm	control	Crestalon 2000 ppm	Leaf drip 1000 ppm	
limonene	0.06	0.05	–	0.08	0.02	0.07	0.05
geyrene	2.23	1.35	2.69	2.92	3.00	3.10	2.55
nonene	4.62	3.94	3.68	4.60	4.02	4.11	4.16
undecene	1.41	1.06	1.57	2.91	2.05	2.33	1.89
anthracene	1.21	1.84	1.77	0.95	1.12	1.13	1.34
neophytadiene	0.07	0.08	0.11	0.20	0.11	0.09	0.11
3,4-dihydrobenzo[b]fluora nthene	1.42	1.91	1.86	1.24	1.28	1.30	1.50
total hydrocarbon compounds	11.02	10.23	11.68	12.90	11.60	12.13	11.59
2-octanone	0.22	0.34	0.30	0.45	0.61	0.55	0.41
2-nonanone	11.64	8.0	12.20	14.45	13.15	12.20	11.94
teteradecanal	1.36	1.11	1.92	1.93	0.98	1.03	1.39
dodecanal	0.13	0.17	0.09	0.20	0.14	0.35	0.18
2-docanone	0.53	0.61	0.62	0.18	0.55	0.63	0.52
2-undecanone	60.74	63.64	62.42	55.88	57.34	57.28	59.55
2-dodecanone	2.82	1.83	1.21	3.27	3.55	3.90	2.76
9-methyl-10-methylene-tricyclo (4.2.1.1.2.5) decan-9-ol	0.19	0.26	0.20	1.31	1.62	1.50	0.85
1-dodecanol, 3.7.11 -trimethyl	0.13	0.71	0.21	1.16	1.29	1.35	0.81
2-tridecanone	2.25	2.86	1.99	0.95	1.77	1.50	1.89
epiglobulol	0.09	0.08	0.04	0.09	0.05	0.10	0.08

elemol	0.73	1.42	1.00	1.25	1.08	1.14	1.10
2-tetradecanone	0.14	0.11	0.15	0.22	0.28	0.27	0.20
nepetalactol	0.10	0.10	0.09	0.14	0.17	0.23	0.14
ascaridole	0.62	2.02	1.00	1.21	1.15	1.19	1.20
guaiol	0.17	0.14	0.15	0.08	0.20	0.16	0.15
eudesmol	0.25	0.07	0.12	0.13	0.19	0.18	0.16
methyl 4-(1,3-benzodioxol-5-yl) butanoate	0.15	0.12	0.09	0.12	0.20	0.27	0.16
hexadecanal	0.13	0.63	0.45	0.16	0.45	0.20	0.34
(Z)-8-(3,5-dimethyl-4-hydroxyphenyl)-2-octene	0.94	0.24	0.21	0.96	0.96	1.10	0.74
9,12,15-octadecatrienal	0.83	0.83	0.61	0.56	0.93	0.66	0.74
hexadecanoic acid	0.36	0.45	0.33	0.18	0.44	0.51	0.38
3-ethoxy-4-hydroxy-4-(4-methoxyphenyl) cyclopent-2-enone	0.04	0.05	0.10	0.04	0.02	0.07	0.05
9,12,15-octadecatrienoic acid methyl ester	1.97	2.57	2.08	1.37	1.16	1.24	1.73
total oxygenated	86.53	88.36	87.58	86.29	88.28	87.61	87.44
total identified	97.55	98.59	99.26	99.19	99.88	99.74	99.04

2-undecanone was identified as the major compound in different treatments and ranged from 55.88% to 63.64%.The second major compound was nonene accounted for 3.68% to 4.62%.This result was in harmony with Pino [27] and Stashenko et al. [28].

Present data revealed that second sowing date caused slightly increased total hydrocarbon compounds, while in the main constituent (2-undecanone) a pronounced decrement was recorded. Application of foliar fertilizers treatments showed inconsistent effect on various compounds of herb essential oil. The trend opposite to that reported with undecanone was observed with nonene, in which second sowing date slightly increased this compound compared to the first one. The highest mean value of 2-undecanone was observed as a result of the interaction between first sowing date and useof Crestalon at 1000 ppm followed by the interaction between the same sowing date and Leaf drip at 2000 ppm. These results were coinciding with those of Khalil [23] on *Rosmarinus officinalis*, Youssef et al. [29] on *Pelargonium graveolens* as well as Khalil and EL-Sherbeny [12] on three *Mentha* species.

CONCLUSION

In general, from the above-mentioned results it would be concluded that sowing rue plants at the end of November was more favorable for production of herb and in addition for more accumulation of essential oil than that of October.

Crestalon and Leaf drip significantly promoted most of studied characters. Leaf drip was more effective in these characters improvement. We may recommend the application of 2000 ppm of Leaf drip to obtain the highest active constituents of essential oil: rutin and coumarin.

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WPŁYW TERMINU SIEWU I STOSOWANIA NAWOŻENIA DOLIŚCIOWEGO NA PLON I SKŁAD CHEMICZNY ZIELA RUTY ZWYCZAJNEJ (RUTA GRAVEOLENS L.)

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Streszczenie

Badanie przeprowadzono w dwóch następujących po sobie sezonach: 2004/2005 i 2005/2006 w celu zbadania wpływu siewu w dwóch terminach i stosowania dwóch różnych nawozów dolistnych na plon i skład chemiczny ruty zwyczajnej (*Ruta graveolens* L.). Przeniesienie siewu z 20 października na 20 listopada spowodowało znaczny wzrost suchej masy ziela (g/m^2), a także plonu i zawartości olejku eterycznego. Z kolei akumulacja zawartości rutyny i kumaryny była wyższa u roślin wysianych wcześniej. Głównym składnikiem olejku eterycznego w zielu jest 2-undekanon. Jego zawartość w olejku eterycznym wyniosła 60,74% u roślin wysianych w pierwszym terminie i spadła do 55,88% u tych wysianych w drugim terminie.
Oba nawozy dolistne: Crestalon i Leaf drip, spowodowały zwiększenie suchej masy ziela, przy czym skuteczność preparatu Leaf drip była wyższa. Generalnie najwyższą zawartość olejku eterycznego, rutyny i kumaryny zanotowano przy zastosowaniu Leaf drip w dawce 1000 i 2000 ppm. Najwyższy plon ziela i olejku eterycznego otrzymano z roślin wysianych w drugim terminie i nawożonych preparatem Leaf drip w dawce 2000 ppm.

Słowa kluczowe: ruta zwyczajna, nawozy dolistne, data siewu, olejek eteryczny, rutyna, kumaryna