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Effect of calcium nitrate applications on leaf macro element concentrations in different strawberry cultivars

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ABSTRACT
Strawberry production is one of the most important horticultural sectors in the Mediterranean coastal line of Turkey. This industry has developed day by day due to high price, yield and good marketing opportunity. However, there are some serious problems about yield due to plant nutrition. The aim of this study was to determine the effects of the calcium nitrate applications on the strawberry leaves nutrition amount during the active growing period (April and May). The results showed that, N and Ca concentrations of the leaves were not affected by different calcium nitrate applications except the period of April. The differences in terms of nutrient contents between cultivars are stemmed from genetical differences. Competition of cation was determined clearly with different doses of calcium nitrate applications and among cultivars.

Keywords:
Calcium
Nitrate
Nutrient uptake
Fragaria x ananassa
Duch

Farklı çilek çeşitlerinde kalsiyum nitrat uygulamalarının yapraktaki makro element konsantrasyonlarına etkisi

ÖZET
Ülkemizin Akdeniz sahilindeki çilek üretimi en önemli bahçe bitkileri sektörlerinden birisidir. Bu üretim alanı; iyi pazarlama olanakları, yüksek fiyat ve verimden dolayı gün geçtikçe artmaktadır. Fakat verimi önemli düzeyde etkileyen bitki beslemeyle ilgili sorunlar vardır. Bu çalışmanın amacı; aktif yetiştirme süresince çilek yapraklarındaki besin element içerikleri üzerine kalsiyum nitrat uygulamalarının etkilerinin belirlenmesidir. Çalışma sonucunda yaprakların N ve Ca içerikleri farklı kalsiyum nitrat dozlarından Nisan dönemi dışında etkilenmemiştir. Çeşitler arasında besin içerikleri bakımından gözlenen farklılıklar büyük oranda genetik farklılıktan kaynaklanmaktadır. Katyon rekabeti, çeşitler arasında ve farklı dozlarda yapılan uygulamalarda açık bir şekilde belirlenmiştir.

Anahtar Sözcükler:
Kalsiyum
Azot
Besin alımı
Fragaria x ananassa
Duch

1. Introduction

Strawberries which grown in many agricultural areas of the world are mainly cultivated in the northern hemisphere. Cultivated strawberries (*Fragaria x ananassa* Duch.), appeared by natural crossing between octoploid *F. chiloensis* (L.) Duch. and *F. virginiana* Duch. species during middle of 18th century (Staudt, 1989). These strawberries have been successfully grown in the temperate areas (Hancock, 1999). Demand to the cultivated strawberry has been increasing continuously in Turkey. Production of strawberry increased from 35.000 tonnes in 1986 to 376.070 tonnes in 2014. Production increase of strawberries in USA, the biggest producer in the world, was 197% between 1986 and 2014; 975% increase occurred in Turkey at the same period (FAO, 2017).

Recently, strawberries became very important due to rapid turn around of investment in and their benefits on human health. Strawberries attract more attention of consumers with mesmerizing aromas, rich vitamin content, especially ellagic acid content and different consumption characteristics (Kafkas et al., 2007). Strawberries' nutritional content depends on cultivation techniques, varieties, fertilization, ecological conditions, and level of fruit maturity. Amount of plant nutrients taken from the soil is controlled by different factors, such as soil type and structure, environmental and biotic factors. Precipitation, temperature and cultural application as well as different soil properties such as soil pH, lime content, amount of organic material and available elements have effect on nutrient uptake. Plant

growth and health are the main biotic factors taken into account. For example age of plant, development situation, cultivar characteristics, structure and health of root system collectively effect nutrient uptake from soil (Erdal et al., 2006).

Like others fruit varieties, it is necessary to meet nutritional demands of strawberries in a timely manner to obtain healthy plants with good yield and high quality fruits. Strawberry varieties show different reactions to applied fertilization programs. Deficiency or excess of plant nutrients may cause different problems during the plant development and fruit maturation. Plant nutrients have a different mobility in xylem, while some of the plant nutrients have high mobility such as nitrogen (N), potassium (K), phosphorus (P), magnesium (Mg), chlorine (Cl) and the others have low mobility in xylem like calcium (Ca) and manganese (Mn) (Karaman, 2012). Calcium affects root elongation (Marschner and Richter, 1974) and cell division, calcium deficiency results in slow down of cell division and root formation. Calcium also maintains hardness and strength of the cell walls by linkage with pectin chains (Kacar and Katkat, 2009).

Fertilizers applied through soil and leaves and their interaction within plants effect overall fruit quality. Wojcik and Lwandowski (2003) reported that application of Ca and B increased these elements concentration in foliage of strawberry cultivar 'Elsanta'; however, these elements had no effect on total and marketable fruit yield, average fruit weight, total soluble solids and titratable acidity. Singh et al. (2007) reported that the calcium applications significantly affect fruit quality by increasing fruit color and reducing grey mould. Palencia et al. (2010) studied the interactions between plant nutrients and occurrence of tip burn, and found no relationship between tip burn and calcium levels. However it was reported that K:Ca and K:Mg ratios were important for the appearance of tip burn symptoms. If these ratios exceed 1.77 for K: Ca and 3.40 for K: Mg, risk of tip burn occurrence would increase by 50%.

In the present study, five strawberry cultivars were used to evaluate effects of different calcium nitrate doses, applied on leaves, on some macro element concentrations in strawberry leaves.

2. Materials and Method

2.1. Plant materials

The study was carried out in a glasshouse at Cukurova University, Adana, during 2012-2013 growing period. Five strawberry cultivars were used as plant material. Cultivar 'Osmanlı' has soft fruit firmness, Camarosa has hard strong fruit firmness and Sevgi, Ebru, and Kaşka cultivars have medium fruit firmness. Calcium nitrate was used as a calcium source in the study.

2.2. Method

Fresh pot plants were planted in a glasshouse on 28th September in fall. Study was set up as randomly split plot experiment design with three replications, and each replication contained 20 plants, where fertilization treatments were the main plots and genotypes were the subplots. Soil was analyzed before the planting and fumigated with Metam sodium and then plants were planted to the fumigated soil within raised beds covered with black polyethylene mulch. Plug plants were planted double row system and designed by triangle method. The distance between the double rows was 1.2 m while the distance between nearby plants in the double row was 0.25 m. Plants were irrigated with drip irrigation system, the common fertilization program (120 kg nitrogen (N) ha^{-1}, 70 kg phosphorus pentoxide (P_2O_5) ha^{-1}, 220 kg potassium oxide (K_2O) ha^{-1} and 20 kg magnesium oxide (MgO) ha^{-1}) was carried out with drip irrigation. Foliar calcium nitrate fertilization was started on 15 January (first blooming time) and continued until 15 May (end of the active vegetative growth period) in 2013. Different calcium nitrate ($Ca(NO_3)_2$) doses (0 (control), 150 , 300 and 450 ml 100 L^{-1}) were sprayed on the leaves with one week intervals, totally 16 times.

Change in nutrient concentrations in leaves were investigated beginning from the blooming date (15th January) until end of the active vegetative growing date (15th May) with one month intervals. Initial leave nutrition concentration was determined in the 15th January for observing the effects of application. Young leaves (10-15) were collected from each treatment, washed and dried at 65^{o}C until reaching the stable weight. Macro nutrient analyses were conducted by dry burning method. The nitrogen concentration was determined via Kjheldahl method. Phosphorous concentration was measured with a spectrophotometer (Dasgan et al., 2008). K, Ca and Mg concentrations were determined with atomic absorption spectrophotometer (Varian brand FS 220 model) with appropriate standards.

2.3. Soil properties

The selected soil physical and chemical properties were determined before planting date. Soil properties were as follows: soil texture was clay loam; $CaCO_3$ amount was 24.6% with a pH of 7.8 (1:2.5 H_2O); EC was 0.31 mmhos cm^{-1}; organic matter was 2.9%; extractable K 193.4 ppm; extractable P 21.20 ppm; extractable Ca 1848 ppm; extractable Mg 121.4 ppm and extractable Fe, Zn, Mn, Cu as 4.23, 1.33, 4.64, 1.48 ppm, respectively.

2.4. Data analysis

All data were subjected to the analysis of variance (ANOVA) using the statistical software package JMP

v.5.0.1 for Windows (SAS Institute, Cary, NC, USA). Differences between means were separated by LSD multiple range tests with a significance threshold of 95%.

3. Results and Discussion

Results for leaf macro nutrient concentrations in different three months were given in Tables 1-3. Results for February and March were not given because they did not differ significantly.

Leaf nutrition concentration in January before calcium nitrate applications were presented in Table 1. While N values were different significantly among the cultivars, P, K, Ca and Mg amounts were not different significantly.

The lowest N content was found in Camarosa (3.65%) while the highest N concentration was found in Kaşka (4.49%) in January. Ebru, Osmanlı and Sevgi had the same statistical group with Kaşka, their N concentrations were 4.43%, 4.19% and 4.18%, respectively.

Table 1. Macro nutrition concentrations (%) in the strawberry cultivars in January

Nutrient	Cultivars					$LSD_{cultivar}$
	Osmanlı	Sevgi	Ebru	Kaşka	Camarosa	
N	4.19 a (1)	4.18 a	4.43 a	4.49 a	3.65 b	0.506*
P	0.28	0.22	0.30	0.34	0.32	N. S.
K	1.36	1.39	1.22	1.36	1.07	N. S.
Ca	2.18	1.95	1.97	2.08	2.13	N. S.
Mg	0.48	0.45	0.46	0.42	0.51	N. S.

(1): Differences of between the means were showed with different letters, N.S.: Non Significant *:$p<0.05$

Rosli et al. (2004) and Serçe et al. (2008) reported that Camarosa variety had high fruit firmness. Nitrogen has a high mobility inside plants, while the vegetative growth increases with high N concentration, fruit firmness and other quality properties can negatively be affected (Turan and Horuz, 2012). At this study, the lowest N concentration was found in the Camarosa variety as known for high firmness, thus it can be suggested that N might have effect on fruit firmness. May and Pritts (1990) reported that 2% - 2.8% N contents were enough for strawberry plants. Nitrogen concentration observed in January in the present study was found to be higher from other published results (Demirsoy et al., 2010; Seferoglu and Kaplan, 2010). It is known that young leaves have higher N concentration than the old leaves, which can explain the results obtained in January measurements (Kacar and Katkat, 1998).

May and Pritts, (1990) determined the level of P concentration between 0.15% and 0.20% in strawberry. Also, Ersoy (2004) determined P concentration as 0.30% in field grown strawberries. In this study, P concentration was found to be between 0.28% and 0.34% in the studied cultivars except Sevgi with a value of 0.22%. Camarosa had the highest Mg concentration (0.51%), at the same time it had the lowest K concentration (1.07%). Plants take Mg up from the soil as Mg^{+2} ions, and K^+ and NH_4^+ ions adversely affect Mg uptake from the soil (Kurvits and Kirkby, 1980).

Macro element concentrations in April were given the Table 2. While the highest concentration of N and Mg was determined in Osmanlı in April, it also had the lowest K concentration.

Similarly, while Camarosa had the highest Ca concentration, it had the lowest concentrations of N, P and Mg. Cultivar and application interactions were determined as statistically not significant for this month.

Phosphorus concentration was affected from applications and seasonal changes, and was found similar to results reported by Almaliotis et al. (2002). But, P concentration decreased in April compared to January.

Demirsoy et al. (2010) report that P concentration of leaves decreased from November to March, but P concentration to increase in May. Phosphorus is heavily used during blooming and fruit development stage (Kacar, 1984). Secondary flowering is high in April month, and it can cause P mobilization from leaves to other parts of the plants thus decreasing overall concentration of P in the leaves.

Calcium concentration of leaves increased with increasing doses of Ca treatments compared to previous months' Ca values. Lieten and Misotten (1993) reported that level of calcium was the highest at blooming and ripening stages due to the increased temperature and intensity of light. They were also reported that the majority of calcium accumulation occurred in leaves and shoots of the plants. Similarly, the highest Ca value

Table 2. The effects of different doses of $Ca(NO_3)_2$ on macro element concentration (%) strawberry cultivars in April

Plant Nutrient	Doses	Cultivars: Osmanlı	Sevgi	Ebru	Kaşka	Camarosa	Average of doses
N	0	4.89	4.48	4.56	4.55	4.52	4.60
	1	4.71	4.57	4.52	4.55	4.38	4.55
	2	4.78	4.62	4.36	4.51	4.44	4.54
	3	4.72	4.53	4.42	4.38	4.41	4.49
Average of cultivar		4.77 a (1)	4.55 b	4.47 b	4.50 b	4.44 b	
$LSD_{cultivar}$**= 0.162		LSD_{dose} = N. S.		$LSD_{cultivar\ x\ dose}$= N. S.			
P	0	0.25	0.26	0.29	0.25	0.25	0.26
	1	0.29	0.25	0.30	0.21	0.24	0.26
	2	0.27	0.25	0.28	0.25	0.26	0.27
	3	0.29	0.25	0.29	0.25	0.21	0.26
Average of cultivar		0.27 ab	0.25 bc	0.29 a	0.24 c	0.24 c	
$LSD_{cultivar}$ ***= 0.023		LSD_{dose} = N. S.		$LSD_{cultivar\ x\ dose}$= N. S.			
K	0	1.36	1.42	1.48	1.76	1.52	1.51 a
	1	1.10	1.36	1.08	1.42	1.36	1.27 b
	2	1.17	1.39	1.44	1.63	1.44	1.42 a
	3	1.18	1.65	1.39	1.47	1.50	1.44 a
Average of cultivar		1.20 c	1.46 b	1.35 b	1.57 a	1.46 b	
$LSD_{cultivar}$***= 0.110		LSD_{dose}***= 0.099		$LSD_{cultivar\ x\ dose}$= N. S.			
Ca	0	3.47	3.31	3.21	3.39	3.70	3.41 b
	1	3.30	3.07	3.48	3.56	3.66	3.41 b
	2	3.60	3.41	3.79	3.42	3.48	3.54 b
	3	3.91	3.61	3.92	3.91	4.10	3.89 a
Average of cultivar		3.57 a	3.53 b	3.60 a	3.57 a	3.74 a	
$LSD_{cultivar}$*= 0.218		LSD_{dose}***= 0.195		$LSD_{cultivar\ x\ dose}$= N. S.			
Mg	0	0.47	0.48	0.44	0.44	0.42	0.45 a
	1	0.48	0.44	0.43	0.41	0.43	0.44 ab
	2	0.46	0.44	0.44	0.39	0.39	0.42 b
	3	0.46	0.41	0.42	0.40	0.38	0.41 b
Average of cultivar		0.47 a	0.44 b	0.43 bc	0.41 bc	0.40 c	
$LSD_{cultivar}$***=0.028		LSD_{dose}*= 0.025		$LSD_{cultivar\ x\ dose}$= N.S.			

(1): Differences of between the means were showed with different letters

N.S.: Non Significant ***:p<0.001; **:p<0.01; * :p<0.05

was found in April when light intensity and temperatures started to increase. Interaction of nutrients were also examined, and it was observed that Osmanlı had the highest Mg (0.47%) and the lowest K (1.20%) concentration.

Agüero and Kirschbaum (2015) also reported negative interaction between potassium and calcium at the different strawberry cultivars.

Macro element concentrations of strawberry varieties treated with different $Ca(NO_3)_2$ doses were given for month of May in Table 3.

N, Ca and K concentrations of the leaves did not change by applications of increasing calcium nitrate doses. However, Mg concentration decreased, which it might be result of cation competition. Nitrogen concentration of leaves decreased compared to April but plants had still sufficient N levels in their leaves. Demirsoy et al. (2010) were determined decrease at the nitrogen concentration of whole plant (leaf, stem and root) at the harvesting period due to the fruit development and transfer to fruits of N.

Phosphorus values of the cultivars in May changed between 0.19% and 0.23%, the lowest P concentrations were measured during growing period. As previously mentioned, plants use P during blooming and fruit development stages, and in return P is mobilized from leaves to flowers and fruits (Kacar, 1984). The lowest K concentration was determined in May during the sampling period. Demirsoy et al. (2010) reported that K concentration of leaves decreased at fruiting stage, K concentration of leaves was the lowest in May. Similar results were also observed in our study.

Table 3. The effects of different doses of $Ca(NO_3)_2$ on macro element concentration (%) in the strawberry cultivars in May

Plant Nutrient	Doses	Cultivars					Average of dose
		Osmanlı	Sevgi	Ebru	Kaşka	Camarosa	
N	0	3.75	3.40	3.52	4.82	3.32	3.76
	1	3.68	3.63	3.41	3.79	3.55	3.61
	2	3.71	3.69	3.73	3.79	3.73	3.73
	3	3.84	3.64	3.46	3.61	3.20	3.55
Average of cultivar		3.74	3.59	3.53	4.00	3.45	
$LSD_{cultivar}$= N. S.		LSD_{dose} = N. S.		$LSD_{cultivarxdose}$= N. S.			
P	0	0.17	0.18	0.20	0.19	0.19	0.19 b(1)
	1	0.26	0.21	0.22	0.18	0.20	0.22 a
	2	0.24	0.19	0.20	0.17	0.20	0.20 ab
	3	0.25	0.18	0.24	0.20	0.21	0.22 a
Average of cultivar		0.23 a	0.19 c	0.22 ab	0.19 c	0.21 bc	
$LSD_{cultivar}$**= 0.023		LSD_{dose}*= 0.021		$LSD_{cultivarxdose}$= N. S.			
K	0	1.20 bcd	1.10b-e	1.38 bc	1.35 bcd	1.19 b-e	1.25
	1	1.11b-e	1.08 cde	1.31 bcd	1.24 bcd	1.03 de	1.16
	2	0.69 f	1.06 cde	1.22 bcd	1.37 bc	1.33 bcd	1.14
	3	0.87 ef	1.14 b-e	1.12 b-e	1.74 a	1.42 ab	1.26
Average of cultivar		0.97 c	1.09 bc	1.26 b	1.43 a	1.24 b	
$LSD_{cultivar}$***= 0.164		LSD_{dose} = N. S.		$LSD_{cultivarxdose}$*= 0.328			
Ca	0	4.05 a	1.98cde	1.85 de	2.12cde	1.89 de	2.33
	1	2.87 b	2.11cde	1.83 de	2.09cde	1.68 e	2.16
	2	2.29 cd	2.26 cd	2.03cde	2.29 cd	2.34 bcd	2.24
	3	2.05cde	2.35bcd	2.08cde	2.46 bc	1.72 e	2.13
Average of cultivar		2.81 a	2.18 bc	1.95 cd	2.24 b	1.91 d	
$LSD_{cultivar}$***= 0.264		LSD_{dose} = N. S.		$LSD_{cultivarxdose}$***= 0.528			
Mg	0	0.43 a	0.34 bcd	0.30 cde	0.30 cde	0.28 de	0.33 a
	1	0.40 ab	0.37 abc	0.26 de	0.26 e	0.27 de	0.31 ab
	2	0.38 abc	0.28 de	0.27 de	0.25 e	0.26 de	0.29 b
	3	0.42 ab	0.30 cde	0.25 e	0.34 bcd	0.12 f	0.29 b
Average of cultivar		0.41 a	0.32 b	0.27 cd	0.29 bc	0.23 d	
$LSD_{cultivar}$***= 0.043		LSD_{dose} = N. S.		$LSD_{cultivarxdose}$*= 0.086			

(1): Differences of between the means were showed with different letters

N.S.: Non Significant, ***:p<0.001; **:p<0.01; *:p<0.05

While P, Ca and Mg concentrations in Osmanlı were higher than that of the other cultivars, its K concentration was lower from the other cultivars. Also Mg concentration of Osmanlı cultivar was the highest with 0.41%.

4. Conclusion

As a well-known phenomenon, plant nutrients are needed at different plant development stages In general, some of the nutrients are in mutual relations between each other. In this study, N and Ca doses of leaves was not affected by different doses of calcium nitrate applied by weekly interval except the period of April. Main reason of differences of the nutrients concentration in the leaves arisen from the cultivars. Competition between the cation was clearly determined with different doses of application and among these cultivars. Macro nutrients change was evaluated in all growing period. It was showed decreasing at leaves due to forming fruit and bloom. The reason of this happen is transportation from leaves to fruit and bloom which are center of food intake.

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