

Genetic variability for mineral concentration of *Eruca sativa* L. and *Diplotaxis tenuifolia* L. accessions

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Received 16 December 2008

Accepted 02 October 2009

ABSTRACT - *Eruca sativa* L. (rocket or arugula) and *Diplotaxis tenuifolia* L. (perennial wall-rocket), are important leafy vegetables and are significant sources of minerals for human nutrition and commonly found in the Mediterranean basin, southern Europe, and Central Asia. The objectives of this study were to determine genotypic variability among and within *E. sativa* and *D. tenuifolia* genotypes for NO_3 , NO_2 , N, P, K, Ca, Mg, Na, Fe, Cu, Zn, and Mn concentrations; to estimate genotype \times environment interaction; and to assess relationships among leaf mineral concentration during two consecutive spring seasons. *E. sativa* and *D. tenuifolia* leaves contained significant amounts of nutritionally important minerals. In general, genotypic variation was lower than phenotypic variation for all mineral concentrations considered, indicating the influence of environment on the expression of analyzed traits. The variance between genotypes and relative importance within genotype variation indicates that NO_3 , NO_2 , K, Mn, Zn, and Cu concentrations may be improved by selecting among cultivars, if the heritability is adequate.

Key words: *Eruca sativa*, correlation, *Diplotaxis tenuifolia*, genetic variance, mineral composition.

INTRODUCTION

Brassicaceae is a large family including many economically important vegetable crops such as cabbage, cauliflower, and broccoli. Genus *Eruca* (sometimes also encompassing the genus *Diplotaxis*) and *Diplotaxis* (Bennett et al. 2006) are minor crops of this family. *Eruca sativa* (rocket or arugula) and *D. tenuifolia* (wild, perennial or sand rocket) are mainly distributed in the Mediterranean region but they also have been cultivated in southern Europe and Central Asia since ancient times (Pignone 1997). The stronger taste of *Diplotaxis* leaves makes them an ideal condiment to add to salads and cooked vegetables. Besides culinary uses, rocket is also considered a medicinal plant (stimulant, antiscorbutic, stomachic, and diuretic oilseed crop) with many reported properties since Roman times (Pignone 1997). *Diplotaxis tenuifolia* is widely

cultivated in Italy (Garibaldi et al. 2004) and found endemic in most Mediterranean countries in Northern and Eastern Europe. Wild rocket is native to the southern and western parts of Turkey but not cultivated nor used as a vegetable.

During recent years consumption of fruits and vegetables has received increased recommendations based on epidemiological studies (Bennett et al. 2006). In some modern cultures, wild plants are still consumed as food and leafy vegetables hold an important place in well-balanced diets (Kawashima and Valente-Soares 2003). Kuhnlein (1990) indicated that leafy green vegetables are significant nutritional sources of minerals. Kawashima and Valente-Soares (2003) reported that raw rucola (*E. sativa*) leaves contain 363 mg K, 5 mg Na, 47 mg Ca, 18 mg Mg, 0.5 mg Fe, 0.4 mg Mn, 0.04 mg Cu, and 0.33 mg Zn in 100 g fresh samples.

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Due to the high glucosinolates content of Brassica vegetables, there is increased interest in cruciferous vegetable breeding for functional foods. Jirovetz et al. (2002) reported that *E. sativa* contains several isothiocyanates and numerous butane, hexane, octane, and nonane derivatives. These compounds constitute the characteristic aroma of the food plant. Many researchers have been interested in sufficient dietary intake of essential nutrients and to increase the consumption of several health promoting compounds by improving plant nutritional properties, nutrient composition and concentration (Bouis 1996, Welch et al. 1997). Enhancing the nutrient concentration of many plant food products would contribute significantly to human nutrition and health (Grusak and DellaPenna 1999). In recent years breeding strategies have focused on the improvement of vitamin and mineral nutrition particularly Fe, Zn, Se and Iodine (Welch and Graham 2004).

Determination of variability in germplasm collections for crucial characters is required for genetic crop improvement (Anshebo et al. 2004). Only a few research studies have focused on genetic variation within vegetables species for mineral accumulation (Kopsell et al. 2004). Significant genotypic variation was reported for minerals, vitamins and other volatile compounds in several species. Wang and Goldman (1996) reported that folate concentration varied four fold among red beet cultivars. Seiler and Campbell (2004) found that wild Jerusalem artichoke genotypic variance made up a significant proportion of the total variation for N, K, P, Ca, Mg, and that the Ca/P ratio was due to genotype, indicating the possibility of improvement through hybridization and selection. These investigations showed that significant variation in nutritional composition already exists within the germplasm of red beet, snap bean, and Jerusalem artichoke and probably exists among many other crop accessions.

Leafy vegetables accumulate significant amounts of nitrates and nitrate itself is relatively non-toxic but its metabolites may produce a number of health effects. Nitrate and nitrite accumulation can be important due to the nitrification process. Nitrite might be accumulated from NO₃ after ingestion, causing methaemoglobinemia (Wright and Davison 1964). However a more recent investigation argues that dietary nitrate may have some beneficial effects, such as antimicrobial effects on gut pathogens (Dykhuzin et al. 1996). Wild rocket

accumulates significant amounts of nitrates and as much as other leafy vegetables such as lettuce and rocket (Santamaria et al. 1995, 1996). The highest nitrate accumulating vegetable is rocket, which absorbs NO₃ and its concentration in leaves can be much higher than in the growth medium (Santamaria 2006).

The primary objective of plant breeding programs over the last several decades has been to increase yields. In recent years researchers are interested in improving the nutritional quality of plants, with respect to both nutrient composition and concentration (Bouis 1996, Kopsell et al. 2004). Concentration is dependent on phenotypic or genotypic variation. Understanding trait inheritance and heritability is important in designing breeding programs (Grant et al. 2008). There is some information about the mineral composition of *E. sativa* and *D. tenuifolia* but no information is available about the genetic variability and heritability of mineral composition of these two species and the potential for breeding for improved mineral content.

The objectives of this study were i) to determine the genotypic variability among and within *Eruca* and *Diplotaxis* genotypes for NO₃, NO₂, N, P, K, Ca, Mg, Na, Fe, Cu, Zn, and Mn concentrations in leaves, ii) to estimate genotype x environment interaction and iii) to examine relationships among mineral composition in rocket salad.

MATERIAL AND METHODS

Twenty seven genotypes were used during two consecutive summer seasons in 2005 and 2006. Twenty accessions were collected from different regions of the world and received from the North Central Regional Plant Introduction Station Ames, Iowa, USA and two *Diplotaxis* genotypes were collected from the Aegean and south-west parts of Turkey (where many *Diplotaxis* species naturally occur) by the authors. One *Eruca* and two *Diplotaxis* genotypes were donated by Dr. Jules Janick, USA, seeds of one *E. sativa* Mill. cultivar were bought and *E. cappadocica* Izgin cultivar which is mainly cultivated for oily seeds, was also obtained from Konya province in central Anatolia in Turkey (Table 1). These different species were evaluated together because the rocket salad name includes different species of *E. sativa* Mill., synonym *E. vesicaria* (L.) Cav., called cultivated rocket. Mild flavored types and pungent types are *D. tenuifolia* well known as wild

rocket, and these species are used for similar purposes and most consumers know *E. sativa* and *D. tenuifolia* as a rocket salad and that it is grown in the same conditions as the same species in many countries.

Experiments were conducted at Ege University, Faculty of Agriculture, Department of Horticulture in Bornova, Izmir, Turkey, latitude 38° 28' N, longitude 27° 15' E, altitude 25 m asl, in the 2005 and 2006 growing seasons. Seeds were sown in February in both years in a 20 liter volume and 75 x 26 x 21 cm size pots, containing a mixture of peat and perlite (3:1) as growth media. Pots were placed in a polyethylene covered glasshouse until the appearance of the first true leaves, then pots were taken to an open field. Randomized complete block design with three replications were used. An individual

plot consisted of one pot and each pot contained 15 plants. No fertilizers were applied and weeds were controlled mechanically by hand and plants irrigated every day. One thousand three-hundred five samples (15 plants x 3 replicates x 29 genotypes) were analyzed for mineral content each year.

A single harvest was done and leaf samples were collected at week 6 after planting. 7-8 leaves were harvest from each plant with stems at least 1 cm above the cotyledons to avoid damaging the apex. Leaf samples were harvested by hand with a knife in the afternoon, due to the leaves showing a much lower concentration of nitrates compared to the morning harvest (Pimpini and Enzo 1996). The total amounts of NO₃, NO₂ and N in the leaf samples were determined by

Table 1. List of genotypes used for the determination of mineral composition

Species (botanical name) ^a	Donor ^b	Accession number/ genotype name/ collection number	Collection locale/ country of origin
<i>Diplotaxis tenuifolia</i> (L.) DC	EUFA-RC	<i>Diplotaxis tenuifolia</i> - I	Antalya-Isparta yolu Turkey
<i>Diplotaxis tenuifolia</i> (L.) DC	EUFA-RC	<i>Diplotaxis tenuifolia</i> - II	Denizli-Acipayam Turkey
<i>Diplotaxis tenuifolia</i> (L.) DC	FRC	<i>Diplotaxis tenuifolia</i>	Italy
<i>Diplotaxis</i> spp.	FRC	<i>Diplotaxis</i> spp.	Italy
<i>Eruca sativa</i>	USDA-ARS	PI 170362	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 173902	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 175720	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 178901	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 179279	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 183233	Egypt
<i>Eruca sativa</i>	USDA-ARS	PI 217829	Pakistan
<i>Eruca sativa</i>	USDA-ARS	PI 251490	Iran
<i>Eruca sativa</i>	USDA-ARS	PI 255664	Afghanistan
<i>Eruca sativa</i>	USDA-ARS	PI 261629	Spain
<i>Eruca sativa</i>	USDA-ARS	PI 311742	Poland
<i>Eruca sativa</i>	USDA-ARS	PI 344365	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 390143	Pakistan
<i>Eruca sativa</i>	USDA-ARS	PI 407630	Turkey
<i>Eruca sativa</i>	USDA-ARS	PI 426198	Afghanistan
<i>Eruca sativa</i>	USDA-ARS	PI 432339	Cyprus
<i>Eruca sativa</i>	USDA-ARS	PI 603033	Pakistan
<i>Eruca sativa</i>	USDA-ARS	PI 633202	England
<i>Eruca sativa</i>	USDA-ARS	PI 597835	Algeria
<i>Eruca sativa</i>	USDA-ARS	PI 633210	China
<i>Eruca sativa</i>	FRC	<i>Eruca sativa</i> - I	Local cultivar- Italy
<i>Eruca sativa</i> Mill.	LC	<i>Eruca sativa</i> - II	Local cultivar- Turkey
<i>Eruca cappadocica</i>	LF	<i>Eruca cappadocica</i>	Konya-Turkey

^a According to ARS-GRIN sample seed package information

^b EUFA-RC: Ege University Faculty of Agriculture, Turkey researchers collected, ARS-GRIN: United States Department of Agriculture Agricultural Research Service North Central Regional Plant Introduction Station, Ames, Iowa, USA, FRC: Received from foreign researchers Jules Janick collected in Italy, LF: Local farmer supported, LC: Local cultivar sales in market

the modified Kjeldahl method, phosphorus (P) in wet digested samples with colorimetry, potassium (K), calcium (Ca) and sodium (Na), with flame photometry and magnesium (Mg), iron (Fe), Zinc (Zn), copper (Cu) and manganese (Mn) using atomic absorption spectrometry (Hills and Jones 1996). Appropriate calibration controls (calibration curve method with commercial certified ICP multi-element standard solution Merck) were applied to each set of measurements. Variance components of cultivars, year and cultivar interaction and error were estimated from the mean squares using an analysis of variance (Becker 1984, Seiler and Campbell 2004, 2006). The phenotypic variance (σ^2_p) was calculated using the following equation:

$$\sigma^2_p = \sigma^2_g + \sigma^2_{gy}/Y + \sigma^2_{ge}/YR$$

where, variance due to genotypes (σ^2_g), the interaction of year and genotype (σ^2_{gy}), standard error (σ^2_{ge}) were calculated from the mean squares of the ANOVAs, while R and Y represent the number of replications and year, respectively as cited in Baye and Becker (2005) (Wricke and Weber 1986, Hill et al. 1998). Estimation of cultivar variance was determined for NO_3 , NO_2 , N, P, K, Ca, Mg, Na, Fe, Cu, Zn, Mn. An ANOVA was conducted for all cultivars within a year using individual plant data (Seiler and Campbell 2006). In order to determine relationships

between mineral nutrition, Pearson correlation coefficients were calculated for individual genotype data from both years. Data analysis was performed with SPSS software (SPSS 16.0).

RESULTS

There were significant differences among the genotypes for all components except Zn in the first experimental year and K, Ca, Zn, and Na in the second year (Table 2). There were also genotypes and year interactions (G x Y) across years and non significant differences were observed for NO_3 , NO_2 , K, Zn, and Mn concentration of *E. sativa* and *D. tenuifolia* genotypes.

The mean and range of values for the *E. sativa* and *D. tenuifolia* mineral element concentration of each genotype for 2005 and 2006 are given in Table 3. The highest NO_3 and NO_2 values were obtained from PI 217829 in both years. The highest P from PI 251490, the highest Fe was obtained from *E. sativa* cultivars. The highest Mg in the first year was from PI 603033 but in the second year it was found in PI 603033 and PI 633210. Ca was highest in PI 178901, the most Cu from PI 603033, and the most Zn from *D. tenuifolia*-II in both years. The highest Mn concentration was from the PI 311742 genotype collected from Poland (Table 4).

Table 2. Analysis of variance components for mineral composition in *E. sativa* and *D. tenuifolia* genotype from the cross-years ANOVA

Mineral element	Statistical significance of mean squares				Variance components ^a				
	Genotype		Cross years	G x Y ^a	σ^2_g	σ^2_{gy}	σ^2_e	σ^2_p	σ^2_g/σ^2_p
	2005	2006							
NO_3	* b	*	** c	ns ^d	149.63	35.40	24.06	165.44	0.90
NO_2	*	**	**	ns	0.00	0.00	0.00	0.00	0.77
N		*	*	*	0.06	0.23	0.02	0.15	0.40
P	*	*	*	*	0.00	0.02	0.00	0.01	0.02
K	*	ns	*	ns	0.56	0.39	0.01	0.69	0.81
Ca	*	ns	*	*	0.00	0.14	0.00	0.05	0.05
Mg	*	*	*	*	0.01	1.37	0.00	0.47	0.02
Na	**	ns	*	*	0.00	0.13	0.00	0.04	0.06
Fe	*	*	**	**	383.09	3845.09	3.14	1665.31	0.23
Cu	**	**	**	**	5.61	0.79	0.01	5.88	0.96
Zn	ns	ns	ns	ns	43.29	6.36	0.46	45.48	0.95
Mn	*	*	**	ns	78.53	10.82	0.48	82.22	0.96

^a G x Y = Genotype x year interaction effects.

^b * Indicates significance at the $P = 0.05$ level of probability based on F-test

^c ** Indicates significance at the $P = 0.01$ level of probability based on F-test

^d NS = Not significant

^a σ^2_g = genotypic variance, σ^2_{gy} = interaction of year and genotype variance, σ^2_e = error variance; $\sigma^2_p = \sigma^2_g + \sigma^2_{gy}/Y + \sigma^2_{ge}/YR$ = phenotypic variance

Table 3. Means and range of values of genotype for mineral composition in *E. sativa* and *D. tenuifolia* genotypes

Mineral element	2005				2006				Overall
	\bar{X}	σ	Min	Max	\bar{X}	σ	Min	Max	
NO ₃ (mg kg ⁻¹)	346.02	13.10	315.92	375.54	318.33	14.48	289.37	346.28	332.18
NO ₂ (mg kg ⁻¹)	0.03	0.00	0.03	0.04	0.03	0.00	0.03	0.03	0.03
N (mg g ⁻¹)	4.40	0.70	2.41	6.20	3.81	0.56	2.07	4.55	4.11
P (mg g ⁻¹)	0.47	0.07	0.24	0.55	0.44	0.05	0.20	0.50	0.46
K (mg g ⁻¹)	6.73	0.88	3.95	8.91	3.99	0.65	2.65	5.77	5.36
Ca (mg g ⁻¹)	1.20	0.16	0.90	1.58	0.94	0.13	0.69	1.26	1.07
Mg (mg g ⁻¹)	0.63	0.04	0.54	0.76	0.50	0.04	0.43	0.61	0.57
Na (mg g ⁻¹)	0.35	0.13	0.16	0.81	0.32	0.08	0.17	0.58	0.33
Fe (mg kg ⁻¹)	127.70	38.01	93.11	277.76	104.45	11.95	81.71	136.83	116.08
Cu (mg kg ⁻¹)	8.42	2.56	3.00	17.01	7.01	2.15	2.45	14.23	7.72
Zn (mg kg ⁻¹)	50.92	7.12	38.13	67.82	38.43	5.98	27.74	52.44	44.68
Mn (mg kg ⁻¹)	53.42	9.56	37.60	72.62	39.12	8.06	25.86	55.19	46.27

The phenotypic and genotypic variance and the ratio of genotypic to phenotypic variation are given in Table 2. The genetic variances for NO₃ among cultivars for NO₂, N, P, Ca, Mg, Na, were very low. The ratio of σ^2_g/σ^2_p was very low for P, Ca, Mg, Na, and Fe; however, higher than 0.77 for NO₃, NO₂, K, Cu, Zn, and Mn. In general, the genotypic variation was lower than the phenotypic variation for all the mineral concentrations considered, indicating the influence of environment on the expression of analyzed traits. Among the mineral concentrations, NO₃ and NO₂ content showed the highest significant and positive correlation coefficient. In addition NO₃ and NO₂ concentration observed similar trends with other nutrient elements except for Fe concentrations (Table 5). Fe concentration was not correlated with NO₃ and positively correlated with NO₂ concentration. P, Mg, and Na were negatively correlated with NO₃ and NO₂ concentrations, whereas NO₃ and NO₂ showed positive associations with N, K, Ca, Cu, Zn and Mn concentrations. These results suggest that high NO₂ and NO₃ content might be accompanied with high N, K, Ca, Cu, Mn, Zn content of *E. sativa* and *D. tenuifolia*. N observed positive associations with K, Ca, Fe, Cu, Zn, Mn, whereas negative associations with Mg and Na concentration. P was positively correlated with Mg, Na, and Cu concentration. Furthermore K showed a highly positive correlation with NO₃, NO₂, N, Ca and a moderate positive relationship with Fe, Zn, Mn but negative associations between P, Na and Mg. The Ca concentration was negatively related with Na, P and Mg while positively correlated with NO₃, NO₂, N, K, Mn, Zn, Fe, and Cu. Likewise Na was highly positively

correlated only with Mg concentration ($r = 0.87^{**}$). Furthermore Mn was positively correlated with NO₃, NO₂, N, K, Ca, Fe, Cu, Zn concentration and negatively correlated with Mg and Na concentration. Zn was positively correlated with NO₃, NO₂, N, K, Ca, Fe and Cu whereas negatively associated with Mg and Na, and not correlated with P concentration.

DISCUSSION

Enhancing nutrient composition of plant food products could contribute significantly to human nutrition and health (Grusak and Dellapenna 1999). Plant breeding has been used to improve crop quality, by increasing concentrations of desirable mineral elements and reducing some potentially harmful elements. However, determination of variability in the germplasm collections for crucial characteristics is required for the genetic improvement of crops for this trait (Anshebo et al. 2004). Wild rocket (*D. tenuifolia*) accumulates a large amount of nitrates with as much nitrates as other leafy vegetables such as lettuce and rocket as our results and other reports have shown (Santamaria et al. 1995, 1996). The large amount of nitrates in both species might be beneficial in the future because dietary nitrate may have some beneficial activity, such as antimicrobial effects on gut pathogens (Dykhuzin et al. 1996).

Potassium accounts for 5% of the total mineral content of the human body and it is a very important mineral to remain healthy. The results revealed that leaf K concentration had a very broad range from 2.65 to 8.91 mg g⁻¹ and a mean of 5.36 mg g⁻¹. Kawashima and

Table 4. Mean of mineral concentration of *E. sativa* and *D. tenuifolia* genotypes

Genotype	NO ₃		NO ₂		N		P		K		Ca		Mg		Na		Fe		Cu		Zn		Mn	
	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006
<i>D. tenuifolia-I</i>	341	312	0.03	0.03	6.1	4.3	0.43	0.41	7.3	4.41	1.3	1.04	0.72	0.58	0.80	0.42	83	10.7	8.9	59	44.8	61	45.5	
<i>D. tenuifolia-II</i>	321	294	0.03	0.03	3.0	2.6	0.53	0.49	7.3	4.40	1.4	1.14	0.64	0.51	0.49	0.45	118	12.2	10.2	67	51.7	54	39.9	
<i>D. tenuifolia</i>	348	321	0.03	0.03	4.0	3.5	0.45	0.42	6.6	3.84	1.3	1.06	0.60	0.48	0.33	0.31	117	7.6	6.3	49	37.1	45	32.3	
<i>Diplotaxis</i> spp.	345	317	0.03	0.03	5.1	4.5	0.43	0.41	5.9	3.28	1.1	0.88	0.67	0.54	0.32	0.30	125	7.6	6.3	44	32.7	40	27.9	
PI170362	338	311	0.03	0.03	4.3	3.8	0.48	0.45	7.3	4.40	1.2	0.94	0.66	0.53	0.37	0.35	112	9.8	9.1	42	30.8	38	26.2	
PI173902	346	317	0.03	0.03	4.7	4.1	0.46	0.43	6.1	3.44	1.2	0.90	0.63	0.51	0.36	0.33	104	7.6	6.3	39	28.1	42	29.4	
PI175720	337	310	0.03	0.03	3.7	3.2	0.52	0.48	6.1	3.44	1.1	0.88	0.55	0.44	0.29	0.27	123	7.6	6.3	61	46.7	57	42.3	
PI178901	338	310	0.03	0.03	4.4	3.9	0.25	0.24	6.7	3.92	1.6	1.24	0.58	0.46	0.26	0.25	111	9.8	6.3	40	29.5	48	34.3	
PI179279	360	332	0.04	0.03	4.7	4.2	0.26	0.24	6.1	3.44	1.1	0.88	0.60	0.52	0.25	0.24	100	8.8	9.1	53	40.6	44	31.1	
PI183233	352	324	0.03	0.03	4.6	4.0	0.50	0.46	6.5	3.76	1.2	0.93	0.65	0.52	0.32	0.30	114	9.9	10.7	89	62	47.7	60	44.7
PI217829	370	341	0.04	0.03	4.3	3.7	0.48	0.45	5.7	3.12	1.2	0.93	0.61	0.49	0.28	0.26	118	9.3	9.1	7.6	44	32.7	44	31.1
PI251490	360	332	0.04	0.03	4.1	3.6	0.54	0.50	6.5	3.76	1.2	0.95	0.61	0.49	0.29	0.27	104	9.1	9.1	7.6	44	32.7	44	31.1
PI255664	332	305	0.03	0.03	3.6	3.2	0.46	0.43	6.1	3.44	1.3	1.04	0.58	0.46	0.48	0.44	145	12.5	7.6	6.3	48	35.6	51	36.7
PI261629	345	317	0.03	0.03	4.1	3.6	0.51	0.47	7.1	4.24	1.2	0.95	0.61	0.49	0.27	0.26	120	10.4	10.7	8.9	51	38.5	69	51.9
PI311742	346	318	0.03	0.03	4.8	4.2	0.48	0.45	8.0	5.04	1.2	0.90	0.60	0.48	0.27	0.26	116	10.1	7.6	6.3	58	44.0	72	54.4
PI344365	346	318	0.03	0.03	4.8	4.2	0.50	0.47	8.0	5.05	1.0	0.75	0.67	0.54	0.38	0.36	125	10.9	7.6	6.3	48	36.1	62	46.3
PI390143	345	318	0.03	0.03	4.8	4.2	0.49	0.46	7.1	4.24	1.0	0.80	0.62	0.50	0.37	0.36	131	11.4	7.6	6.3	53	40.1	55	40.7
PI407630	351	323	0.03	0.03	5.1	4.5	0.45	0.43	7.3	4.41	1.1	0.88	0.66	0.53	0.39	0.36	120	10.4	3.0	2.5	41	30.0	65	48.7
PI426198	343	315	0.03	0.03	4.6	4.0	0.46	0.43	6.9	4.08	1.1	0.83	0.57	0.46	0.29	0.28	129	11.2	3.0	2.5	44	32.6	54	39.9
PI432339	328	300	0.03	0.03	4.7	4.2	0.41	0.39	7.1	4.24	1.1	0.88	0.63	0.51	0.39	0.37	112	9.8	7.6	6.3	47	34.8	48	34.3
PI597835	368	339	0.04	0.03	3.7	3.2	0.45	0.42	6.9	4.08	1.2	0.90	0.62	0.50	0.27	0.26	123	10.7	9.0	7.6	58	44.0	63	47.1
PI603033	349	321	0.03	0.03	5.1	4.5	0.51	0.47	6.1	3.44	1.0	0.80	0.75	0.60	0.38	0.36	116	10.1	16.8	14.0	54	40.9	70	53.6
PI633202	363	334	0.04	0.03	4.8	4.2	0.49	0.46	7.0	4.24	0.9	0.70	0.59	0.47	0.32	0.30	120	10.4	7.6	6.3	49	37.1	45	31.9
PI633210	362	334	0.04	0.03	4.7	4.1	0.50	0.47	7.8	4.88	1.0	0.75	0.65	0.52	0.34	0.32	120	10.4	6.1	5.1	48	35.6	60	44.7
<i>E. sativa-I</i>	350	295	0.03	0.03	4.4	4.0	0.47	0.47	8.8	5.69	1.2	1.11	0.62	0.47	0.29	0.27	141	13.5	7.6	5.1	52	39.8	67	34.3
<i>E. sativa-II</i>	331	303	0.03	0.03	2.4	2.1	0.52	0.48	4.0	2.69	1.3	1.06	0.61	0.49	0.16	0.17	274	13.2	9.1	7.6	61	46.9	52	38.3
<i>E. carpoudecica</i>	359	330	0.03	0.03	4.9	4.3	0.49	0.45	6.3	3.60	1.5	1.19	0.66	0.53	0.23	0.23	121	10.6	10.7	8.9	51	38.5	55	40.7

Valente-Soares (2003) made similar observations of K concentration and reported that *E. sativa* leaves contained 3.63 mg g⁻¹ and Cavarianni et al. (2008) reported 4.3-5.5 mg g⁻¹. Leafy green vegetables are excellent sources of many nutrients, particularly Ca which is very significant in human diets. Ca concentration of *E. sativa* and *D. tenuifolia* were affected by genotype and values were similar in both years. Ca concentration depended on genotype and changing mean value was 1.07 mg g⁻¹ which is a close result to what Cavarianni et al. (2008) reported (2.3-3.1 mg g⁻¹) in different rocket salad cultivars grown in Nutrient Film Techniques. Cultivar variations in leaf Ca concentration among collards have been reported in response to soil or foliar applied Ca fertilization (Johnson 1991). Differences in climate, soil, agronomic practice, location and plant cultivar are the probable causes for differences in nutrient levels (Kawashima and Valente Soares 2003).

Mg concentration varied among species and genotype such as wild rye (Jefferson et al. 2001), wheatgrass (Mayland and Asay 1989) and broccoli (Mark et al. 2000). In this study, Mg concentration varied among genotypes with an average concentration of 0.57 mg g⁻¹. Kawashima and Valente-Soares (2003) reported that *E. sativa* Mg concentration was 0.30 mg g⁻¹, and Cavarianni et al. (2008) 0.43-0.46 mg g⁻¹ which are very similar to our findings. Mineral concentrations can differ across tissues within a single plant, across genotypes of a given species, or more broadly across species (Arzani et al. 2007). Fe is an important mineral element

and mainly found in large amounts in spinach, and has immune-stimulant effects like Zn and Fe nutrients (Bukshs et al. 2007). Agte et al. (2000) suggested that leafy green vegetables have good potential as a natural supplement for Fe and Zn. Correlation analyses between 12 mineral concentrations showed that Mn and Ca concentrations were significantly positively correlated with Zn concentration as Jiang et al. (2007) reported in milled rice (*Oryza sativa* L.). Furthermore Hacisalihoglu et al. (2005) found that in faba bean seed the concentration of Zn showed a positive relationship with Fe, Cu and Mn, with similar trends observed among present investigation.

When nutrient concentration was assessed in various plants, significant genotypic variation was observed for minerals. The variance between genotype and relative importance of within genotype variation indicate that genotype may be used to improve NO₃, NO₂, K, Mn, Zn, and Cu concentrations by selecting among cultivars. Farnhem et al. (2000) indicated that phenotypic variation was determined among commercial and inbred lines of broccoli for both Ca and Mg concentration. Harrison and Bergmann (1981) showed significant differences for leaf P, Ca, Mn, Cu, and Zn concentrations in three cabbage cultivars. Bukshs et al. (2007) indicated that nutrient element compositions may vary due to botanical structures of plant and mineral compositions of soil. Karamanos et al. (1994) reported mineral composition of the faba bean may vary due to genotype variation, collected location and environmental conditions. Results of the rocket salad

Table 5. Pearson correlation coefficients for mineral composition in *E. sativa* and *D. tenuifolia* genotypes based on combined data for two years

Mineral element	NO ₃	NO ₂	N	P	K	Ca	Mg	Na	Fe	Cu	Zn
NO ₂	0.969**										
N	0.744**	0.848**									
P	-0.235**	-0.251**	-0.345**								
K	0.603**	0.713**	0.892**	-0.229**							
Ca	0.666**	0.791**	0.914**	-0.334**	0.828**						
Mg	-0.676**	-0.799**	-0.939**	0.355**	-0.821**	-0.934**					
Na	-0.771**	-0.865**	-0.867**	0.281**	-0.756**	-0.877**	0.872**				
Fe	0.072	0.183*	0.288**	0.039	0.292**	0.434**	-0.394**	-0.331**			
Cu	0.233**	0.249**	0.261**	0.203**	0.126	0.308**	-0.294**	-0.243**	-0.030		
Zn	0.443**	0.538**	0.631**	-0.027	0.610**	0.707**	-0.715**	-0.597**	0.391**	0.539**	
Mn	0.511**	0.563**	0.637**	-0.008	0.656**	0.590**	-0.592**	-0.589**	0.261**	0.308**	0.660**

** Indicates significance at the $P = 0.01$ levels of probability

* Indicates significance at the $P = 0.05$ levels of probability

nutrient composition indicate that the origin of the collected material may influence the investigated nutrient composition. Most papers reported similar findings and observed genetic variation in mineral concentration and Di Giacomo et al. (2007) reported the content of mineral and trace elements is a good instrument for establishing a geographical place of origin of Fucino potatoes and that they were variety grouped potatoes collected from origin. Rodriguez Galdon et al. (2008) applied multivariate analysis and principal component analysis data on mineral and trace elements in onion bulbs from various seed origins. Chope and Terry (2009) evaluated different onion cultivars for mineral composition and indicated significant differences in the mineral composition of the bulb due to the genotypic variation. For the investigated minerals, genotypic variation was lower than phenotypic variation, indicating the influence of environment on the expression of the analyzed traits. A similar observation was reported by Anshebo et al. (2004) in sweet potatoes. Hacısalihoglu et al. (2005) reported a significant variation for mineral nutrients and indicated greater genetic variation in bean lines for Zn, Fe, Mn, Cu, P and K concentration. The present study indicated a high phenotypic variance for σ^2g/σ^2p ratio of NO_3 , NO_2 , K, Mn, Zn, and Cu concentration of *E. sativa* and *D. tenuifolia*. The minerals N, P, Ca, Na, Mg, Fe, gave the lowest σ^2g/σ^2p ratio. Natural variation occurs for both essential and nonessential trace elements in crop species and in cultivars within species

(Bell et al. 1997, Huang and Graham 2001, Grant et al. 2008). Seiler and Campbell (2004) found that in wild Jerusalem artichoke, genotypic variance components accounted for a significant proportion of the total variation for N, K, P, Ca, and Mg, and that the Ca/P ratio was due to genotype and indicated the possibility of improvement through hybridization and selection. Kuhnlein (1990) indicated that leafy green vegetables are significant nutritional sources of minerals. Improvement of the nutritional quality of our food supply, especially with respect to essential nutrient minerals, such as Mg, Fe and Zn, could be an important goal of leafy vegetable crop breeding.

In conclusion the present study represents the survey of genetic variation for several nutrient compositions in rocket salad. Furthermore results indicate that *E. sativa* and *D. tenuifolia* genotypes observed rich nutritional concentration and that there is genetic variability for the different mineral composition among genotypes even though the environment accounted for a great variation as well. Additional studies on heritability are required if a selection is to be used for the improvement of mineral concentrations in *E. sativa* and *D. tenuifolia*.

ACKNOWLEDGEMENTS

The authors thank North Central Regional Plant Introduction Station, Iowa, USA for providing seed samples and research funds of the Ege University, which supported a part of this study.

Variabilidade genética nas concentrações minerais em *Eruca sativa* L. e *Diplotaxis tenuifolia* L.

RESUMO - *Eruca sativa* L. e *Diplotaxis tenuifolia* L. são vegetais importantes para a nutrição humana como fonte de minerais e podem ser encontrados comumente nas regiões mediterrâneas, no Sul da Europa e na Ásia Central. O objetivo deste estudo foi determinar a variabilidade genotípica para as concentrações de NO_3 , NO_2 , N, P, K, Ca, Mg, Na, Fe, Cu, Zn e Mn, entre e dentro de genótipos de *E. sativa* e *D. tenuifolia*; estimar a interação genótipo x ambiente; e avaliar a relação entre as concentrações foliares desses minerais durante dois cultivos consecutivos de primavera. As folhas de *E. sativa* e *D. tenuifolia* apresentaram quantidades significativas de minerais nutricionalmente importantes. Em geral, a variação genotípica foi menor que a variação fenotípica para todas as concentrações dos minerais considerados, indicando a influência do ambiente sobre a expressão dos caracteres analisados. As variações entre os genótipos e a relativa importância das diferenças genotípicas indicam a possibilidade de sucesso com a seleção entre os cultivares em relação às concentrações de NO_3 , NO_2 , K, Mn, Zn e Cu, se a herdabilidade for apropriada.

Palavras-chave: *Eruca sativa*, correlação, *Diplotaxis tenuifolia*, variação genética, composição mineral.

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