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Accumulation of Nitrate in the Roots of Carrot in Conditions of Transylvania and Correlation Between the Amount of Soluble Dry Matter and Nitrites

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Abstract. Nitrate accumulation in plants is a natural phenomenon resulting from uptake of nitrate ion in excess of its reduction and subsequent assimilation. The problem of nitrate (V) accumulation in vegetable plants is very important to a potential consumer, due to the reported negative impact of nitrates on human health. The aim of this study was to determine the nitrate contents in edible parts of 2 varieties of carrot, before clamping and after uncovering, in fresh carrot and dry matter. Achievement of a rational system of supplementary fertilization requires setting of the most appropriate combination of fertilizers, the ratio between these epochs and the doses of administration and knowledge of how each variety or hybrid reacts to the fertilizers used. Also, if organic carrots are used then the organic fertilizer has natural nitrates unlike the chemical fertilizer which can create a higher concentration of nitrates. The mode of fertilization and its influence on the content in nitrates shows that organic fertilization leads to accumulation of smaller quantities of nitrates in carrot roots. Organic fertilization results in a reduction in the nitrates from roots with 1.89 mg/100 g, respectively 1.91 mg/100 g in 2010, and for the year 2011 of 2.47 mg/100 g and 2.68 mg/100 g, as compared with the chemical fertilization.

Keywords: cultivar, carrot root, nitrates, nitrites.

INTRODUCTION

Carrot (*Daucus carota* L.) is the important vegetable crop, grown in temperate climate zone. Consumption of carrot, both fresh and processed, has increasing tendency. Nitrate accumulation is affected not only by the type of nitrate fertilizer used, but also by nitrogen rates, variety, environment, harvesting date and other agronomical factors (Boskovic-Rakocevic *et al*., 2012; Kòňa *et al*., 2006).

The content of inorganic nitrate (NO_3^-) in certain vegetables and fruit can provide a physiologic substrate for reduction to nitrite (NO_2^-), nitric oxide, and other metabolic products (NO_x) that produce vasodilation, decrease blood pressure, and support cardiovascular function (McKnight *et al*., 1999; Lundberg *et al*., 2006).

Nitrate toxicity is manifested by its ability to transform into nitrosamines; nitrosamines can be formed in both food products (exogenous origin), during their preserving, and digestive system (endogenous origin), especially in the stomach (Muresan *et al*., 2012; Santamaria 2006).

Nitrate accumulation in plants is a natural phenomenon resulting from uptake of nitrate ion in excess of its reduction and subsequent assimilation (Gajewiski *et al*., 2009). The relationship for carotenoid and nitrates accumulation was much weaker (below 50%). The relationship between dry matter of the roots and sugars or soluble solids accumulation was also strong (Bajer, 2009).

MATERIALS AND METHODS

The experiments were carried out in 2010 and 2012 in Garbau, Cluj County. In the study were followed three experimental factors: carrots varieties (Flakker 3 and Nantes-5), sowing date (March and May) and fertilization applied (organic – cattle manure and chemical – NPK complex 16:16:16). After harvesting were used for the determination of nitrate content of carrot roots 2 on each repetition. Determination of nitrate content was performed using spectrophotometer.

The spectrophotometric method consisted in nitrite dosing, by measuring colour intensity at 520 nm of the azo formed compound after the reaction of dissolution between the sulphanilic acid and the nitrites from the aqueous extract of the sample and coupled with alpha-naphtylamine. To a new portion from the aqueous extract, nitrates are reduced to nitrites with metalic dmium and the total nitrite content is determined. The difference between the total nitrites content and the original nitrites content are the nitrates (STAS 9065/2002).

RESULTS AND DISCUSSIONS

For root vegetables Ord.M.S. 975/1998 specifies the maximum permissible nitrates content in carrots. The Acceptable Daily Intake (ADI) of nitrate and nitrite set by European Commission's Scientific Committee for Food (ECSCF), is 3.7 mg/kg body weight, and 0.06 mg/ kg body weight, respectively (WHO, 1995).

In the experimental year 2010, cultivar influence on the nitrate content in carrot roots is shown in Tab. 1, where the highest value was recorded at the variety Nantes-5, 14.88 mg/100g sp. Flakker-3 variety has recorded a very significant negative difference of 1.63 mg/100g sp.

As a result of the performed analysis, the nitrate content values varied 70 mg/kg for the samples of Romania carrots.

Tab. 1

The unilateral influence of cultivar contend on nitrates in carrot roots

Cultivar	Year							
	2010				**2011**			
	Nitrates		±D	Signification.of	Nitrates		±D	Signific.of
	mg/100g	%	mg	difference	mg/100g	%	mg	difference
Nantes-5 (Mt)	14.88	100		-	14.51	100	-	-
Flakker -3	13.25	89.0	-1.63	00	12.71	87.59	-1.80	000
LSD 5			0.60				0.90	
LSD 1%			1.39				2.09	
LSD 0.1%			4.43				6.64	

Late sowing (May) leads to a very significant reduction of nitrate content as compared to the early sowing (March), the decrease being of 1.90 mg/100g sp in 2010 and 1.80 mg/100g sp in 2011.

Following the analysis, it can be observed that the values of the nitrate content are lower than the ones obtained by Muresan et al. in 2012, in which case the values were 70 mg / kg for carrot samples from Romania.

Tab. 2

Unilateral influence of the sowing period on nitrates content in carrot roots

Sowing period	Year							
	2010				**2011**			
	Nitrates		±D	Signific.of	Nitrates		±D	Sig of diff
	mg/100g	%	mg	difference	mg/100g	%	mg	
early (Mt)	15.02	100	-	-	14.51	100	-	-
late	13.12	87.3	-1.90	000	12.71	87.5	-1.80	000
LSD 5			0.37				0.41	
LSD 1%			0.54				0.67	
LSD 0.1%			0.81				1.26	

The mode of fertilization and its influence upon the nitrates content shows that organic fertilization leads to the accumulation of a lower quantity of nitrates in carrot roots and as compared to the chemical fertilization it registered a mean reduction of 2.57 mg/100g sp in 2010 and 2.31 mg/100g s.p. in 2011, the differences being very significant in both cases (Tab. 3). Nitrate content in organic carrots was reduced by 20-70% with potassium fertilizer application through leaves compared to carrots on calcium-magnesium nitrate (Zaborskienė *et al.*, 2009).

Tab. 3

The unilateral influence of the mode of fertilization upon the content of Nitrates in carrot roots

Fertilization	Year							
	2010				**2011**			
	Nitrates		±D	Signific.of	Nitrates		±D	Sig of diff
	mg/100g	**%**	mg	difference.	mg/100g	**%**	mg	
chemical (Mt)	15.22	100	-	-	14.90	100	-	-
Organic	12.91	84.8	-2.31	000	12.33	82.7	-2.57	000
LSD 5			0.21				0.34	
LSD 1%			0.35				0.49	
LSD 0.1%			0.65				0.74	

With delaying of harvest term, the content of nitrates in the roots significantly decreased (Gajewiski *et al*., 2009).

By combining the two factors, cultivar and sowing period, presented in Tab. 4, for 2010, as regards the nitrates content in roots, it can be observed that the variety Flakker-3, regardless of the time of sowing, recorded the lowest values of nitrates, the differences being significant as compared to the variety Nantes-5 that is considered the control sample.

In the year 2011, the amount of nitrate values in the varieties Nantes-5 and Flakker-3 was lower than in 2010. Variety Flakker-3, regardless of the period of sowing, recorded lower values, with distinct significant differences (1.68 for early age and 1.58 respectively for the late age), as compared to the variety Nantes-5.

Tab. 5 presents the situation in the case of the interaction of the factors cultivar and the nature of the fertilizer upon the nitrates content in roots, during the period of culture 2010-2011.

Tab. 4

Influence of the cultivar and sowing period on nitrates content in carrot roots

Cultivar	Sowing period	Year							
		2010				**2011**			
		Nitrates		±D mg	Significant of difference	Nitrates		±D mg	Sig of diff
		mg/100g	%			mg/100g	%		
Nantes -5(Mt)	early	15.86	100	-	-	15.43	100	-	-
Flakker-3	early	14.17	88.4	-1.68	00	13.59	88.0	-1.85	0
Nantes -5(Mt)	late	13.91	100		-	13.59	100	-	-
Flakker-3	late	12.33	88.6	-1.58	00	11.84	87.1	-1.75	0
LSD 5				0.68				0.97	
LSD 1%				1.32				2.10	
LSD 0.1%				3.52				6.17	

In 2010, for the variety Nantes-5 the values of nitrate content in roots ranged between 12.29 mg/100 g and 16.23 mg / 100g, the variety Flakker-3 recorded distinct significant negative differences of 2.02 mg/100 g, following chemical fertilization, respectively significant of 1.24 mg/100 g following organic fertilization as compared to Nantes-5 considered control sample.

In the year 2011, is highlighted the variety Flakker-3 in which the nitrate content in roots is less than the content in roots for the variety Nantes-5, with 2.51 mg/100 g following chemical fertilization, the difference being distinctively significant, and significant of only 1,09 mg/100 g, following organic fertilization.

Tab. 5

Influence of the cultivar and fertilization on nitrates content in carrot roots

Cultivar	Fertilization	Year							
		2010				**2011**			
		Nitrates		±D mg	Significant of difference	Nitrates		±D mg	Sig of diff
		mg/100g	%			mg/100g	%		
Nantes -5(Mt)	chemical	16.23	100	-	-	16.15	100	-	-
Flakker-3	chemical	14.21	87.5	-2.02	00	13.64	84.4	-2.51	00
Nantes -5(Mt)	organic	13.53	100	-	-	12.87	100	-	-
Flakker-3	organic	12.29	90.8	-1.24	0	11.78	91.5	-1.09	0
LSD 5				0.63				094	
LSD 1%				1.39				1.99	
LSD 0.1%				4.21				5.87	

Combining the factors sowing period and fertilization (Tab. 6) shows that both in the case of early sowing period and late sowing period, organic fertilization results in a reduction of the nitrates content in roots with 1.89 mg/100 g, respectively 1.91 mg/100 g in 2010, and for the year 2011, of 2.47 mg/100 g and 2.68 mg/100 g, as compared to the chemical fertilization, the difference being very significantly negative.

Following a comparative analysis of all experimental variants, in which the three studied factors interacted, as regards the content of nitrates in carrot roots, during the years 2010-2011 (Tab.7), it is noted that:

- There are statistical assured differences between most variants tested
- The highest content of nitrates, regardless the time of sowing, is recorded at the variety Nantes-5 with chemically fertilized;

- The lowest nitrate content in carrot roots is recorded in the late era of sowing in the variety Flakker-3 organically fertilized (11.13 mg/100g sp) followed by variety Nantes-5 (12.37 mg/100g sp) ;
- late sowing period influenced the low level of nitrate in carrot roots for variety Flakker-3 both organically and chemically fertilized.

Tab. 6

The influence sowing dates and fertilization on content of nitrates carrot in roots

Sowing period	Fertilization	Year							
		2010				**2011**			
		Nitrates		±D mg	Signific.of difference	Nitrates		±D mg	Sig of diff
		mg/100g	%			mg/100g	%		
early (Mt)	chemical	16.17	100	-	-	15.74	100	-	-
early	organic	14.28	88.3	-1.89	000	13.28	84.3	-2.46	000
late (Mt)	chemical	13.87	100	-	-	14.05	100	-	-
late	organic	11.96	86.2	-1.91	000	11.37	80.9	-2.68	000
LSD 5				0.53				0.48	
LSD 1%				0.77				0.70	
LSD 0.1%				1.15				1.04	

Tab. 7

Combined influence ofcultivar, sowing period and fertilization on nitratecontent in carrot roots

Sowing period	Fertilization	Cultivar	Nitrates (mg/100g)	Significance
early	chemical	Nantes-5	17.09	A
early	chemical	Flakker-3	14.82	C
early	organic	Nantes-5	14.20	D
early	organic	Flakker-3	13.11	E
late	chemical	Nantes-5	15.29	B
late	chemical	Flakker-3	13.03	E
late	organic	Nantes-5	12.37	F
late	organic	Flakker-3	11.13	G
DS		0.15-0.16		

For a better interpretation of the results it was necessary to correlate the experimental data, expressing the content in soluble solid content, and the level of nitrites in carrot roots.

The record values of soluble dry matter content in root ranged from 9.24 and 11.63%, these being good for the conditions of the area. Moniruzzaman *et al*., in (2013), obtained slightly higher values.

The maximum dry matter content of root (15.90%) was obtained from N_2 while the minimum dry matter content of root (9.87%) was obtained in control treatment.

From the results, it was found that the increase in the amount of nitrite value is related to increase in the amount of soluble dray matter. In both cultivars studied, were revealed linear equations calculated for the two sowing periods (early - March, late - May), variations to which chemical fertilization was applied. Regarding the variety Nantes-5, for the two sowing periods distinct significant linear correlations are established (r = 0.52 ** - sown in March, r = 0.66 ** - sown in May), between the values of soluble dry matter and the amount of nitrites (Fig. 1).

High levels of the percentages of soluble dry matter of 10.67 (sown in May) and 10.35 (sown in March) lead to achieving high levels of the amount of nitrites (0.35 mg / kg sp – sown in May and 0.34 mg / kg sp – sown in March).

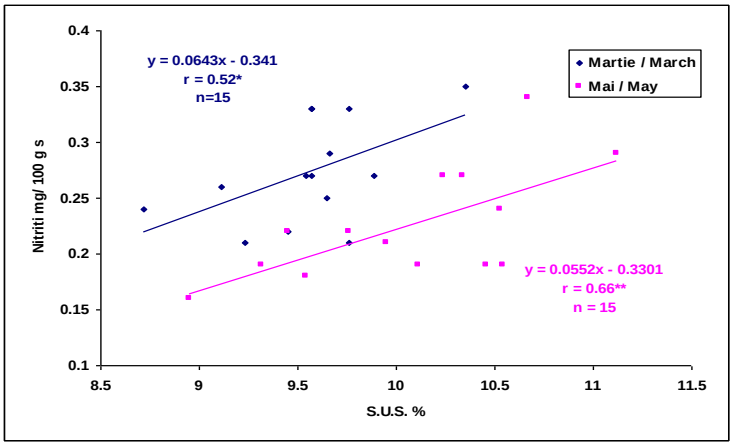

Fig.1.Correlation between the amount of soluble dry matter and of nitrites (mg/kgs.p.) in Nantes-5varieties, chemically fertilized (average2010-2011)

In the case of variety Flakker-3 (Fig. 2), the relationship between the percentage of soluble dry matter and the amount of nitrites is represented by significant distinct linear equations significant for the two sowing periods (r = 0.80 ** - sown in March, r = 0.85 ** - sown May).

Reduced values of the amount of soluble dry matter (9.95% - sown in March, 9.72% - sown in May) determine low values of the amount of nitrites (0.16 mg / kg sp respectively 0.34 mg / kg sp sown in May and March).

Higher values of the correlation coefficient (r = 0.80 and r = 0.85) show a direct stronger link between the content of soluble dry matter and nitrites content in the roots of the variety Flakker-3 chemical fertilized, as compared to the variety Nantes -5.

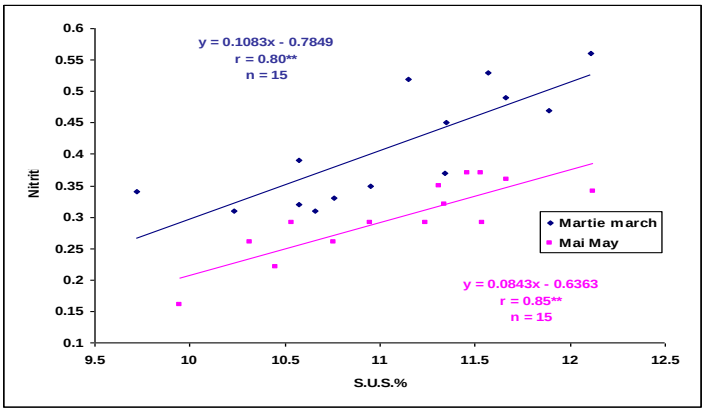

Fig.2.Correlation between the amount of soluble dry matter and of nitrites (mg/kgs.p.) in Flakker-3 variety, chemically fertilized (average 2010-2011)

Regarding organic fertilization, for the variety Nantes-5, the sowing periods are represented by significant distinct linear correlations (r = 0.82 ** in case of early sowing - March and r = 0.83 ** for the late sowing - May) between the amount of soluble dry matter values and the values of nitrites (Fig. 3).

For the cultivar Nantes-5, organically fertilized, high values of nitrites of 0.24 mg / kg sp (sown in May) and 0.31 mg / kg s.p. (sown in May) are obtained in high percentages of soluble dry matter (11.13% respectively 10.34%).

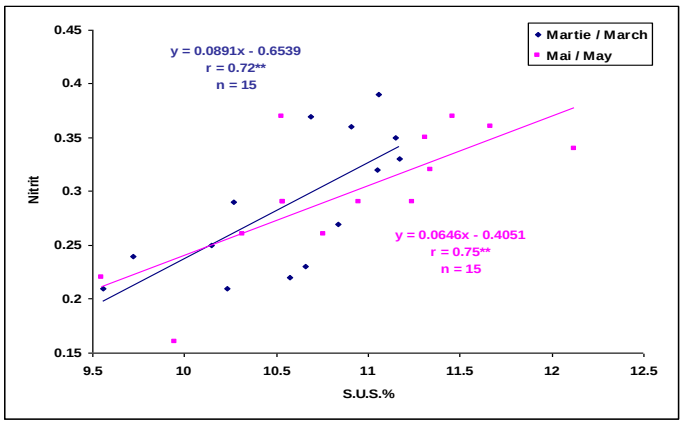

Fig. 3. Correlation between the amount of soluble dry matter and of nitrites (mg/kg s.p.) in Nantes-5 variety, organically fertilized (average 2010-2011)

Regarding the cultivar Flakker-3, organically fertilized, correlating the percentage of the values of dry matter with the level of the nitrites, linear correlations were obtained, distinctly significant (r = 0.72 ** - sown in March and r = 0.75 ** - sown in May) (Fig. 4).

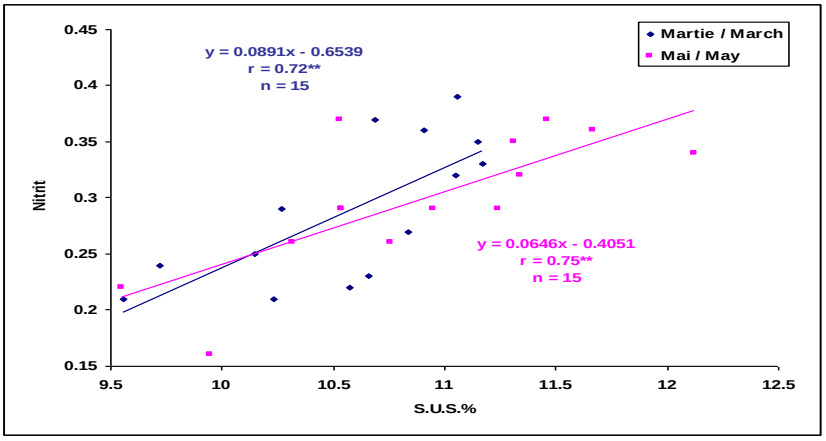

Fig.4. Correlation between the amount of soluble dry matter and of nitrites (mg/kg s.p.) in Flakker-3 variety, organically fertilized (average 2010-2011)

CONCLUSION

The nitrate content in the roots of carrot as limiting factor of the food value varies depending on the variety, growing period and fertilizer applied phasially. The lowest content of nitrates is registered in the variety Flakker- 3, especially in the variant sown late and organically fertilized (11.13 mg/100 g s.p.). In addition, the variety Nantes-5, in the same variant, sown late and organically fertilized, the nitrate content is lower than in the other variants (12.37 mg/100 g s.p.). These values are below the maximum allowed limit by regulations in force 40 mg/100 g s.p.

Higher values of nitrates content are found in carrots from crops sown early and chemical fertilized, especially in the variety Nantes-5 (17.09 mg/100 g s.p.).

The nitrite content in carrot roots is correlated with that of soluble dry matter. This correlation is more significant in the culture of the variety Nantes-5 with organic fertilization

(r = 0.82 to 0.83) and less significant at the same variety with chemical fertilization (r = 0.52 to 0.66).

It is recommended to use organic fertilization for both varieties of carrots, due to the fact that it has recorded the best values and the sowing periods should be before the end of May.

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