RESEARCHES CONCERNING NITRATES AND NITRITES ACCUMULATION IN CARROTS, ALONG OF THE VEGETATION STAGES

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Abstract. The presented paper deals with the determination of nitrates and nitrites content in carrots, in different vegetation stages of the carrot culture. High nitrates and nitrites concentration in vegetables is mainly due to excessive nitrogen content in the soil system, thus deteriorating the nutritional and hygienic values of products and complicating the processing and storage. The determination was tested on carrot samples assayed from an experimental field set up near Timisoara. In experimental field, to the carrot culture was administrated different doses of fertilizers (NPK) and the samples for analysis were assayed in different phases of vegetation. The obtained results indicated that the highest level of nitrate in carrots was found to the variant b_3 ($N_{150}P_{90}K_{90}$) in experimental field, who was above maximum limit allowed (LMA). Maximum limit allowed for nitrates in carrots, in accordance with ORDER No. 293/640/2001-1/2002 regarding security and quality conditions for vegetables and fresh fruits for human consumption is 400 ppm. For all other samples of carrots the nitrates level was below of LMA. The nitrite content grows in case of fertilizer samples in all stages of vegetation. The nitrite content in carrots should not exceed 1-2 ppm. Nitrate and nitrite content in carrots was done with the help of High Performance Liquid Chromatography (HPLC) in the Laboratory for the Measurement of Residues of the Department of Agro-techniques of the U.S.A-V.M.B in Timisoara.

Keywords: carrots, nitrate content, nitrite content, HPLC

INTRODUCTION

Nitrate is a naturally occurring form of nitrogen and is an integral part of the nitrogen cycle in the environment. Nitrate is formed from fertilizers, decaying plants, manure and other organic residues. Due to the increased use of synthetic nitrogen fertilizers and livestock manure in intensive agriculture, vegetables and drinking water may contain higher concentrations of nitrate than in the past. The presence of nitrate in vegetables, as in water and generally in other foods, is a serious threat to man's health. Nitrate per se is relatively non-toxic, but approximately 5% of all ingested nitrate is converted in saliva and the gastrointestinal tract to the more toxic nitrite. The only chronic toxic effects of nitrate are those resulting from the nitrite formed by its eduction by bacterial enzymes [1].

Nitrite as such, and nitrate when reduced to nitrite, may react with amines or amides to form carcinogenic *N*-nitroso compounds. Nitrosation can occur mainly in two situations: **1.** during storage and ripening of the food product; **2.** in the stomach from the action of salivary nitrite produced through enzymatic reduction of endogenous or exogenous nitrate [2]. In accordance with the **Order No. 293/640/2001-1/2002** regarding security and quality conditions for vegetables and fresh fruits for human consumption, maximum limit allowed (LMA) for nitrates in carrots is 400 ppm [3]. The maximum limit allowed (LMA) for nitrite should not exceed 1 - 2 ppm.

MATERIALS AND METHODS

In 2007 have been assayed carrots with the purpose to determinate nitrate and nitrite concentration, in different vegetation stages of the culture. The samples were taken from an experimental field, set up near Timisoara. In the field have been administrated 3 doses of synthetic fertilizers (b1 - $N_0P_0K_0$, b₂ - $N_{90}P_{90}K_{90}$, b₃ - $N_{150}P_{90}K_{90}$) and one dose of rother

fertilizer ($b_4 - 40$ t/ha). Nitrate and nitrite content in carrots was done with the help of High Performance Chromatography (HPLC) in the Laboratory for the Measurement of Residues of the Department of Agrotechniques of the U.S.A-V.M.B in Timisoara.

The HPLC method parameters: Column: C18, IonoSphere A 250 mm; Mobile Phase: 0,01 M Potassium hydrogen Phtalate; Pressure (1 Mpa = 10 bar = 145 psi): 6.3 MPa; Flow rate: 1.2 ml/min; Temperature: 25°C; Sample volume: 30 μ l; Detection: UV at 290 nm; Cell volume: 12.0 μ l.



Figure 1. High Performance Liquid Chromatograph

RESULTS AND DISCUSSIONS

The obtained experimental results are shown in tables 1 & 2.

Carrot samples were assayed when plants had 5-6 leafs in roseda, to the beginning of the root inssipation (after 25-30 days) and to full maturity (when the roots have minimum 1.5 - 2 cm to roseda grosness).

The nitrate and nitrite content accumulated in plants depends of enzymatic transformations that have place along the vegetation phases. The transformations that nitrates it supports in plants, in the first stage, consist in two successive reductions, catalyzed by nitratereduction and nitrite-reduction enzymes. These enzymes contain minor elements (nitrate-reduction enzymes contain molybdenum, and nitrite-reduction enzymes contain lead and cooper).

Variant	NO ₃ (ppm) content in carrots Vegetation phases		
	$b_1 (N_0 P_0 K_0)$	89	79
$b_2 (N_{90}P_{90}K_{90})$	144	126	101
$b_3 (N_{150}P_{90}K_{90})$	417	276	260
b ₄ (manure 40 t/ha)	262	190	121

Table 1. NO₃ content in carrots in different vegetation phases

The nitrates reduction is influenced by the light intensity and by the presence of molybdenum, which provide the needed electrons for the reduction reaction. The less molybdenum quantity, the more is the accumulation of nitrate in plants.

$$NO_3^- \xrightarrow{\text{nitratreduction}} NO_2^- \xrightarrow{\text{nitritreduction}} NH_4^+$$

Figure 2. The nitrogen transformations in plant.

The higher nitrate content in carrot samples was registered to variant b_3 in 5-6 leafs stage, (417 ppm), variant that have been administrated the following dose $N_{150}P_{90}K_{90}$, but along the vegetation phases of the

culture the nitrate content decrease so to the beginning of the root inssipation the nitrate content decrease to 276 ppm and to full maturity reach to 260 ppm.

The maximum limit allowed (LMA) for carrots according with Order No. 293/640/2001-1/2002 regarding security and quality conditions for vegetables and fresh fruits for human consumption is 400 ppm. The exceeding of the maximum limit allowed of 400 ppm was registered to variant on that have been administrated synthetic fertilizer with maximum nitrogen content (N₁₅₀K₉₀P₉₀).

	NO ₂ (ppm) content in carrots			
Variant	Vegetation phases			
	5-6 leafs	Beginning of the root inssipation	Full maturity	
$b_1 (N_0 P_0 K_0)$	1,44	0,84	0	
$b_2 (N_{90}P_{90}K_{90})$	1,47	1,96	2,35	
$b_3 (N_{150}P_{90}K_{90})$	2,28	3,57	4,67	
b ₄ (manure 40 t/ha)	1,47	2	3,5	

Table 2. NO₂ content in carrots in different vegetation phases

The nitrite content grows to those variants on which have been applied NPK inorganic fertilizer, accountable to blind sample, the increase being in direct proportion to applied nitrogen quantity. To the variant b_4 on which we used manure, the nitrite content retrieval in vegetal samples is about equal to the nitrite content from variant b_2 , but much lower than b_3 variant.

The nitrite quantity from vegetable species grows once with plants maturity, as follow of nitrate decomposition in nitrite under the action of nitratereduction enzymes. The decomposition process of nitrite in ammonium ion, and of inorganic fraction in inorganic compounds with low molecular weight is deferred once with full maturity of plants, a possible cause being the small content of cooper and lead in carrot.

The exceeding of normal values (1-2 ppm) for nitrite in vegetable species, was registered to variant on which have been applied nitrogen inorganic fertilizer in proportion of $N_{150}P_{90}K_{90}$. In the case of variant fertilized with manure and nitrogen fertilizer ($N_{90}P_{90}K_{90}$), the nitrite content exceed the 2 ppm limit to full maturity of plants.

CONCLUSIONS

 The nitrate content in analyzed samples decrease during the vegetation stages of plant. The exceed of maximum limit allowed of 400 ppm, was registered only in the case of nitrogen fertilizers administration in variant $N_{150}P_{90}K_{90}$, in 4-5 leafs stage.

- The nitrite content grows in the case of nitrogen fertilizer administration along the vegetation phases. Variant N₁₅₀P₉₀K₉₀ lead to high nitrite content above LMA, in samples, in all vegetation stages, and manure administration get high nitrite values to full maturity of plant.
- The optimum fertilizer variant, which does not decide nitrate and nitrite accumulation in carrots, in the present experimental conditions, is synthetic fertilizer NPK in proportion of 90:90:90.
- The nitric compounds accumulation in plants is realized in different ways depending by the vegetation period and harvesting time, climatic conditions and most by the light intensity, lead to nitrate reduction in plants and the nitrate content diminish in harvested products.

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