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RESEARCH ON THE RESIDUAL LEVEL OF NITRATES IN SOME CHEESE PRODUCTS

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Abstract
The research was done on 40 samples of fresh cow cheese, 20 samples from sheep's milk cheese, 20 samples of cow's milk green ewe cheese, 90 cottage cheese samples and 92 samples of cacciocavallo.

Key words: nitrates, cheese products, green ewe cheese, milk, residues, animals.

INTRODUCTION

For milk and dairy products there has been used the method of cadmium column reduction and expression of the ions was made in ions of NO^-_3 and NO^-_2.
Samples of milk mixture (10 ml) were initially subjected to correct deproteinization after their dilution with hot distilled water and the addition of protein-free substances and filtering to obtain a filtrate volume of 200 ml volume will be considered in the calculation.

MATERIAL AND METHOD

In the next phase, the extract filtered through folded filters and free of NO^-_3 şi NO^-_2, which must be clear, we proceeded to:
- reduction of nitrates to nitrites. A proportion of 20 ml filtrate into the reservoir of cadmium column and buffer solution and the eluate was collected in a 100 ml flask. After filtration of the eluate there were done two successive washings of the tank wall column with 15 ml doubly distilled water and then the tank column was filled with doubly distilled water. The volumetric flask was retired filled to volume eluate and doubly distilled water and was it homogenized.
Calculation of the nitrite in NO^-_2 at a l of milk was established after the relation:

$$\text{ppm}NO^-_3 = \frac{100000 \cdot C1}{V \cdot V1}$$

The level of nitrates of the sample expressed in nitrate ions (NO^-_3) was established at 1 l of milk after the relation:

$$ppmNO_3^- = \left(\frac{100000 \cdot C1}{V \cdot V1} - NO_2^- \right) \cdot 1{,}35$$

The same method was used for sheep's milk, reconstituted milk powder and cheese, indicating that for work with cheese (fresh cottage cheese, green ewe cheese cheese from sheep's milk, cow's milk green ewe cheese) there were used 10 g of the product.

RESULTS AND DISCUSSIONS

For the fresh cow cheese the results are presented in table no.1.
The obtained data show limits of ions NO^-_3/kg between 0.40 to 2.30 mg / kg and an average of 1.23 mg / kg. The average value of nitrates established in cheese is set at less than for cow milk and would represent about 45% of the value for milk.

Table 1

The content of nitrates in fresh cow cheese

Year	No. of samples	mg NO^-_3/kg		Overcome of LMA
		Limits	Average	
2009	20	0,40-2,10	1,245	0
2010	20	0,40-2,30	1,220	0
Total/Average	40	0,40-2,30	1,232	0

The results for the 20 samples of cow's milk cheese and 20 samples from sheep's milk cheese are presented in Tables. 2, 3.

The analysis of obtained data at cow milk cheese showed:

- an average of 1,06 mg NO^-_3/kg of cow milk green ewe cheese in the year 2009 and limits between 0,80 and 1,70 mg NO^-_3/kg;
- an average of 1,02 mg NO^-_3/kg în the year 2010, and limits between 0,60 şi 1,80 mg NO^-_3/kg;
- an average of 1,04 mg NO^-_3/kg at all studied samples in the two years for the cow milk green ewe cheese;
- there have not been found values of the nitrate content in the cow milk green ewe cheese that could infleuence negatively the security of consumers.

Table 2

Content of nitrates in green ewe cheese from cow milk

Year	No. Of samples	mg NO^-_3/kg		Overcome of LMA
		Limits	Average	
2009	10	0,80-1,70	1,06	0
2010	10	0,60-1,80	1,02	0
Total/Average	20	0,60-1,80	1,04	0

Table 3

Content of nitrates in sheep's green ewe cheese

Year	No. of samples	mg NO^-_3/kg		Overcome of LMA
		Limits	Average	
2009	10	0,30-0,90	0,52	0
2010	10	0,10-1,00	0,50	0
Total/Average	20	0,10-1,00	0,51	0

Data on nitrate in sheep milk green ewe cheese shows:
- An average of 0.52 mg NO^-_3/kg of sheep's green ewe cheese, with limits ranging between 0.3 and 0.9 mg NO^-_3/kg in 2009;
- An average of 0.50 mg NO^-_3/kg and limits between 0.1 to 1.00 mg NO^-_3/kg 2010;
- An average of 0.51 mg NO^-_3/kg for all samples investigated. Comparing the data obtained from two kinds of cheese there can be noticed lower values of sheep milk green ewe cheese (0.51 mg NO^-_3/kg) than from cow milk (1.04 mg NO^-_3/kg).

We believe that the decrease in nitrate content of cow's green ewe cheese at the rate of about 61% of the nitrate content of milk and the rate of about 76% in the green ewe cheese from the sheep's milk was due to the shift of a part of nitrates in the whey and metabolism during maturation of another part by some microorganisms.

Data on levels of nitrate content in fresh cow cheese, green ewe cheese from cow's milk and sheep's milk green ewe cheese show low levels of residual nitrate in these products, levels ranging from 0.51 mg to NO^-_3/kg in sheep curd and 1.23 mg NO-3/kg the fresh cow cheese.

For cow milk cottage cheese and / or a mixture of cow and sheep milk, research has been extended for four years at a total of 90 samples. These types of cheese valued by consumers and products to meet market requirements in the area were taken from both production units and the producers from small farms, which exploit the agro-food product markets. The results obtained are shown in Table. 4.

For the entire studied period, the average content of nitrate ions NO^-_3/kg that had a value of 1.51 mg / kg, but limits ranged from relatively large values between 0.40 and 5.00 mg / kg.

For 2007 and 2008, the average values amounted to 1.68 mg and 1.70 mg / kg of NO^-_3, and the limits ranged from 0.50 mg nitrate / kg. Average values were very close and met for the years 2009 and 2010, ie 1.27 mg NO^-_3/kg in 2009 and 1.30 mg NO^-_3/kg in 2010. And in this case the limits of variation were relatively large, between 0.40 mg and 4.10 mg NO^-_3/kg.

The relatively small amounts of nitrate in cheese matured or premature, that after the last salting will be covered with whey and can be explained by:
- Conversion of nitrate to nitrites and harmless products in the process of maturation;
- Elimination of brine after equalization salt concentration in the cheese and whey, except that starting after the 30 days maturation nitrates may increase, as determined by Alexandrina Trif et al. (1996).

Table 4

Average level of residual nitrates in cottage cheese

Year	No. of samples	mg NO^-_3/kg		Overcome frequency of LMA
		Limits	Average	
2007	25	0,50-4,20	1,68	0
2008	25	0,50-5,00	1,70	0
2009	20	0,40-2,80	1,27	0
2010	20	0,40-4,10	1,30	0
Total/Average	90	0,40-5,00	1,51	0

To determine the hierarchical level of nitrate content in the samples investigated in the four years, there has been made a classification of content and distribution of nitrate on the levels of 0, <1, 1.1 to 2, 2.1 to 3, 3.1 -4, 4.1 to 5 mg**.**

The results of this distribution and the calculation made in absolute and relative frequency are shown in the table 5.

The data show the following situation:
- level < 1 mg NO^-_3/kg at 31 samples representing 34,44%;
- level between 1,1-2,00 mg NO^-_3/kg at 39 samples, ie. 43,33%;
- level between 2,1-3 mg NO^-_3/kg at 17 samples, ie. 18,89%;
- level between 4,1-5 mg NO^-_3/kg at 3 samples, ie. 3,33%;
- level o mg NO^-_3/kg and 3,1-4,00 mg NO^-_3/kg has not been found.

Table 5

Absolute and relative frequency of nitrates in cottage cheese according to its content

Content of mg NO^-_3/kg	2007		2008		2009		2010		Total	
	A	R	A	R	A	R	A	R	A	R
0 mg/kg	0	0	0	0	0	0	0	0	0	0
< 1 mg/kg	8	32,00	9	36,00	7	35,00	7	35,00	31	34,44
1,1-2 mg/kg	8	32,00	8	32,00	12	60,00	11	55,00	39	43,33
2,1-3 mg/kg	8	32,00	7	28,00	1	5,00	1	10,00	17	18,89
3,1-4 mg/kg	0	0	0	0	0	0	0	0	0	0
14,1-5 mg/kg	1	4,00	1	4,00	0	0	1	0	3	3,33
Total	25	100	25	100	20	100	20	100	90	100

The analysis of data showed that the rate of nitrate ions was at most cheese samples (62.23%) between 1.1 mg-2, 00 mg NO-3/kg. The content level of

less than 1 mg / kg (<1 mg) were within 34.44% of the samples and the content of 4.1 to 5 mg / kg and 3.33% were placed in evidence.
The processed cheese category in the form of cacciocavallo were investigated in 92 samples for nitrate content in the years 2007-2010. The results obtained are shown in Table. 6.

The obtained data for each year have esteblished the following aspects:

- an average of nitrate ion content of 1,24 mg NO^-_3/kg in the year 2007;
- an average of 1,11 mg NO^-_3/kg in the year 2008;
- an average of 1,00 mg NO^-_3/kg in the year 2009;
- an average of 1,08 mg NO^-_3/kg in the year 2010.

In all investigated samples he average content of nitrate ion / kg had a value had the value of 1.11 mg NO^-_3/kg. The data showed a relatively wide variation in the nitrate limits at this kind of cheese, ranging from 0 mg / kg and 6.80 mg / kg of nitrate ions (NO^-_3).
To highlight the levels of nitrate content after 0 mg / kg, <1 mg / kg, 1-2 mg / kg, 2.1 to 3 mg / kg, 3.1 to 4 mg / kg, 4.1 to 5 mg / kg and 5.1 to 6.8 mg / kg there was established absolute and relative frequency of these NO^-_3/kg ion content.

The obtained data is presented in table 7.

The classificiation of nitrate levels according to the announced criterium shows that:

- in 10 samples (10,87%) the nitrate level was of 0 mg NO^-_3/kg;
- in 42 samples (45,65%) the nitrate levels were situated under 1 mg NO^-_3/kg;
- in 27 samples (29,35%) the nitrates had values between 1-2 mg NO^-_3/kg;
- in 6 samples (6,52%) the nitrates had values between 2,1-3 mg NO^-_3/kg;
- in 7 samples (7,60%) the level of nitrates had been over the value of 3,1 mg NO^-_3/kg.

Table 6

Annual average values of ppm content of nitrate ions in cacciocavallo

Year	No. of samples	mg NO^-_3/kg		Frequency of overcome LMA	
		Limits	Average	A	R
2007	25	0,10-5,00	1,24	0	0
2008	25	0-6,80	1,11	0	0
2009	22	0,10-2,20	1,00	0	0
2010	20	0-3,80	1,08	0	0
Total/Average	92	0-6,80	1,11	0	0

These aspects show that at 79 samples (85,87%) the nitrate content had values between 0 mg and 2 mg NO^-_3/kg, and at 13 samples (14,13%) the nitrates had greater values than 2 mg NO^-_3/kg.

Table 7

Absolute and relative frequency of nitrate levels in cacciocavallo according to their contents

Level of mg NO^-_3/kg	2007		2008		2009		2010		Total	
	A	R	A	R	A	R	A	R	A	R
0	0	0	5	20,00	0	0	5	25,00	10	10,87
<1	14	56,00	10	40,00	12	54,54	6	30,00	42	45,65
1-2	7	28,00	7	28,00	7	31,82	6	30,00	27	29,35
2,1-3	1	4,00	1	4,00	3	13,64	1	5	6	6,52
3,1-4	1	4,00	1	4,00	0	0	2	10,00	4	4,35
4,1-5	2	8,00	0	0	0	0	0	0	2	2,17
5,1-6,8	0	0	1	4,00	0	0	0	0	1	1,09
Total	25	100	25	100	22	100	20	100	92	100

A = absolute frequency; R = relative frequency (%)

Comparing the frequency set at different amounts of nitrate in cottage cheese and cacciocavallo as cheese products that are subject to the maturation process there have been established:

- a content between 0 and 2 mg NO^-_3/kg at 85,87% in samples of cacciocavallo şi of about 78% in cottage cheese;
- values of nitrate content greater than 2 mg NO^-_3/kg there has been met with a frequency of 22,23% at the samples of cottage cheese and with a frequency of 14,13% at the samples of cacciocavallo.

CONCLUSIONS

In cheese products the contents of these NA had values under AML.
In fresh cheese products the average content of nitrates in milk was lower than 1.23 mg NO^-_3/kg values in the fresh cow cheese; 1.04 NO^-_3/kg in the curd from cow's milk and 0.51 NO^-_3/kg from sheep milk green ewe cheese.
In both types of investigated maturated cheese, nitrates had the average values of 1.51 mg NO-3/kg in cottage cheese to 1.11 mg NO^-_3/kg cacciocavallo.
The reduced nitrate content in cheese products represents the following percentages of the nitrate content of milk: 45.55% from fresh cow cheese, 38.52% from cow's milk curd, 23.28% from sheep milk green ewe cheese; 56,60% in cottage cheese and 41,12% in cacciocavallo.
The nitrite content of fresh cow cheese, curd from cow milk and sheep milk green ewe cheese is in much lower than that determined in the milk used.

These redictions were made 2,4 times for the fresh cow cheese ($\bar{x}$ = 0,026 ppm NO^-_2), for about 6,5 times for cow green ewe cheese ($\bar{x}$ = 0,0095 ppm NO^-_2) and for about 10 times for the sheep green ewe cheese. ($\bar{x}$ = 0,0065).

In cottage cheese the nitrite content had limit values between 0 and 0,3 ppm NO^-_2, the average value being 0,0444 ppm NO^-_2, thus more reduced than the one from the milk 1,4 times and in about 68% of the samples there have not been found nitrites.

In processed cheese in the form of cacciocavallo, about 74% of the samples were free of nitrites; in the samples with nitrites these presented limit values between 0,10 ppm and 0,30 ppm NO^-_2 and the average value was of 0,032 ppm NO^-_2 1,9 times lower than the nitrite content of milk.

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REFERENCES

1. Ciocârlie N., 2000, Cercetări privind evidenţierea unor substanţe toxice de contaminare chimică în produsele alimentare de origine animală, în zona de est a ţării. Teză de doctorat USAMV Bucureşti.
2. Mergey C, J. M. Bennoit, 1978, Determination of Nitrate in Food by a Nitrat specific Electrod. Analisis, 6, 164-172.
3. Mîrzac L., 2001, Evaluarea toxicologico-igienică a acidului oxalic şi oxalaţilor în produsele alimentare şi nutreţurilor. Teză de doctorat F.M.V., Chişinău.
4. Rădulescu H., M. Goian, 1999, Poluarea nitrică a alimentelor. Ed. Mirtav, Timişoara.
5. Rotaru O., M. Mihaiu, 2002, Igiena veterinară a produselor alimentare. Patologie prin alimente. Ed. Todeso, Cluj-Napoca.
6. Stănescu V., C. Laslo, C. Guş, 1984, Eficienţa unor metode pentru depistarea $NaNO_3$ şi $NaNO_2$ din lapte. Lucrări Simp. Prob. actuale în controlul alimentelor, 146-151.
7. Trif A., V. Curtui, Gh. Milovan, D. Pârvu, C. Căpăţână, D. Baicu, 1994, Rezidual nitrates and nitrites in pasteurized milk, yoghourt and green cheese preparated from milk with nitrat content over the tolerated limits. Lucr. şt. USAB, Timişoara, 28, 169-173.
8. Trif A., Gh. Milovan, V. Curtui, C. Sala, D. Pârvu, M. Drugă, 1999, Dynamics of nitrate and nitrite a long the preparation process of „Telemea" cheese. Lucr. Simp. International „Integrated Systems for Agrafood Production. Timişoara, 75-82.
9. Trif A., V. Curtui, M. Drugă, G. Suiugan, 2000, Screeningul azotaţilor şi azotiţilor în unele sortimente de brânzeturi comercializate pe piaţa liberă în Timişoara. Lucr. şt., 6, 192-194.
10. 2001, Ord. MAAPM nr. 356.
11. 2001, Ord. MAAPM nr. 357.