

**NITRATES AND NITRITES IN MEAT PROCESSED PRODUCTS  
REGULATION AND OCCURRENCE*****Ilijana Boci<sup>1</sup>, Elda Ziu<sup>2</sup> Marsida Tuzi<sup>2</sup>***<sup>1</sup>*Department of Industrial Chemistry, Faculty of Natural Sciences, University of Tirana*<sup>2</sup>*University of Shkodra, Shkodra, Albania**\* Corresponding author: ilirjana.boci@fshn.edu.al***ABSTRACT**

Use of nitrite and nitrate as common food additives is a controversial issue. Both represent a class of preservatives mostly used during meat processing. Nitrite is an approved additive considered a foremost curing ingredient for the preservation of meat products, nitrate as well. Except for that as nitrate is frequently encountered in the environment, it is considered as a natural constituent of the human diet that raises concern for its potential harmfulness related to the human health conditions and environmental contamination. Nitrite is commonly used in cured meat products while nitrate is used mainly in long shelf life raw fermented meat products. The use of both chemicals is regulated by Albania Directive "Food Additives other than colorants and sweeteners".

The authors of this paper investigated the occurrence of these chemicals in two selected cured meat products because of an ongoing discussion and rumour in local media for presence of nitrate in cured meat products though even well below the allowed maximum level. As all the producers sustain they do not use nitrate potassium/sodium in their cured meat product, the authors analysed two kinds of sausages in different national and foreign laboratories to confirm the presence or not of nitrate and more important, trying to explain the route of nitrate entering in these products. The results of nitrate findings in both products were inconsistent showing fluctuations between labs though all results were well below the maximum limit. The authors tried to explain the cause of this presence by analysing raw material and processing water or considering other ingredients where nitrate would be present as a contaminant. The respective analysis confirmed that nitrate originated from these sources contributed at only a detectable presence of nitrate in cured meat products leading to the conclusion that this persistent but fluctuating nitrate presence is mainly consequence of the chemical biological conversion of initial nitrite to nitrate. Potential factors influencing this conversion should further be studied.

**INTRODUCTION**

One of the most troubling problems, in terms of meat processed products preservation is the use of nitrite and nitrate as multifunction additives. Nitrite and nitrate are used as food additives in cured meat to stabilize the color of red meat, inhibit spoilage and growth of food poisoning organisms and contribute to flavor development. Nitrate is also used in the preservation of fish products and the production of cheese. Since 1995, nitrate and nitrite are listed as officially accepted preservatives in European Union (EU) legislation (European Union, 1995). Nitrite and nitrate are key intermediates of various biochemical reactions in the nitrogen cycle. Nitrate is the most fully oxidized nitrogen compound and therefore stable to oxidation, but potentially a strong oxidizing agent. In hypoxic conditions, nitrate can be reduced and then acts as an oxidizing agent, with release of energy. Nitrite can be oxidized to nitrate by strong chemical oxidants or by nitrifying bacteria or reduced to nitrogen oxides through several enzymatic and non-enzymatic pathways, producing energy. Because of their high bioavailability, nitrite/nitrate are capable of playing complicated and contradictory roles as food additive, natural contaminants and/or contaminant. Emerging studies strengthen the new understanding of the role of nitrate and nitrite in the human body, motivating revision of the long-held view that these ions pose a health risk. (Merino et al, 2017) Research has shown that there are indisputable benefits of nitrite and nitrate in promoting human health, suggesting that these ions could be considered indispensable dietary components and even used as possible therapeutic agents. The scientific debate is ongoing. Here are two sources of nitrate and nitrite in the body: exogenous (external) and endogenous (internal) (Merino et al, 2017). Human exposure to nitrate is mainly from the exogenous source, while exposure to nitrite is mainly endogenous, through nitrate metabolism. The intake of exogenous nitrates and nitrites is mainly via food, particularly vegetables, meat products and drinking water (EFSA, 2008).

The limit for addition of nitrate (E 251 = NaNO<sub>3</sub>, E 252 = KNO<sub>3</sub>; all additive products expressed as NaNO<sub>3</sub>) during processing ranges from 100 to 150 mg/kg for processed meat and cheese products. In addition, the current legislation allows certain traditional products to be produced based on residual amounts, with the maximum residual amount ranging from 10 to 300 mg/kg for traditionally cured meat products (European Union, 2001b).

The statutory limit on the use of nitrite (E 249 = KNO<sub>2</sub>, E 250 = NaNO<sub>2</sub>, expressed as NaNO<sub>2</sub>) in meat products is 150 mg/kg and in heat-treated processed meat 100-150 mg/kg. Maximum accepted residual amount for various traditional products ranges from 50 to 180 mg/kg (European Union, 2011).

**MATERIALS AND METHODS**

The samples taken for analysis were two kinds of sausages that constitute the most common products traded and consumed in domestic market produced from one Albanian meat processing company. Both samples were analyzed for nitrite and nitrate content even though the producers do NOT use nitrate in these cooked meat products, only in raw fermented ones. The samples analyzed by chance from one control laboratory presented a considerable content of nitrate in both samples, except for nitrite (below its MRL). As all the meat producers in Albania sustain not to have used nitrate in their products, we followed this case. The repetition of analyses from the same lab confirmed the nitrate presence but at somewhat lower level (well below the MRL).

We analyzed the same products in our lab and confirmed the presence of nitrate at low level comparable to nitrite content. The literature confirm that the fate of nitrate and nitrite is complex especially in meat processed matrices. Nitrate (NO<sub>3</sub><sup>-</sup>) is the most fully oxidized compound of nitrogen and is therefore stable to oxidation.

Nitrate acts as an oxidizing agent, being itself reduced to nitrite (NO<sub>2</sub><sup>-</sup>) in the process. (Merino et al, 2017). All samples were analyzed for nitrite and nitrate using a spectrophotometric method based on reduction of nitrate with zinc powder developed at the Swedish NFA (Merino 2009). The initial nitrite concentration and total nitrite after Zn reduction are determined based on Griess reaction.

**Figure 1:** Two kinds of sausages analyzed for nitrite/nitrate content

Oxidation of nitrite in nitrate in meat products is not a well known fact to contribute at a considerable amount of nitrate in meat products strangely resulting in some cases at a higher amount than initial nitrite added in the product. Nitrate and nitrite are ions which co-exist in most foodstuffs and actually analysts rarely find one without the other. For this reason, methods for the analysis of nitrite/nitrate are considered together here.

The method used in our lab for determination was according to Determination of Residual Nitrite/Nitrate in Foodstuffs and Water After Zinc Reduction Leonardo Merino Published online: 7 October 2008. The control lab and foreign lab have reported to have used ISO 3091:75 which is based on Cd reduction of nitrate to nitrite.

The aim of this paper is not to make the comparison of the methods used from labs, so all the deviations in results within and between labs have not been considered, because we want to confirm the presence of nitrate or not and to explain why. Both samples analyzed are shown in the Fig.1

**RESULTS AND DISCUSSION**

Literature says that levels of residual nitrite and nitrate in meat products are variable because they depend on the time and temperature used during processing and storing, the initial addition of nitrite and nitrate, the composition of the meat, pH, addition of antioxidant components such as ascorbate and the presence of micro-organisms.

Our results for nitrite and nitrate content for both kinds of samples analyzed are shown in the Figure. 2. Obviously the products are the same type but the time of analysis between the labs is different. Our lab tested the same product in consecutive days.

Product Type 1	NaNO <sub>2</sub> mg/kg	KNO <sub>3</sub> mg/kg
Our lab first day	6.3	36.8
Our lab	5.46	22.5
Our lab	3.33	47.4
Our lab last day	2.55	33
1st control lab last day	6.78	160
2nd control lab last day	<2	<5
Product Type 2	NaNO <sub>2</sub> mg/kg	KNO <sub>3</sub> mg/kg
Our lab first day	74.55	11.24
Our lab	58.5	13.3
Our lab	47.01	29
Our lab last day	28.5	96
1st control lab last day	14.52	100.9
2nd control lab last day	58.4	72.2

**Table 1:** Nitrite and nitrate content found in two kinds of sausages

Other than nitrite values that are decreasing obviously because of the time dependence as expected, the table below shows a more meaningful and interesting information regarding the nitrate content in both samples even if no nitrate is added as additive in these products.

Although now, some scientists are emphasizing the beneficial effects of dietary intake of nitrate after the discovery that nitric oxide (derived from nitrate and nitrite) plays an essential role in the immune system, the problem of this nitrate presence in meat processed products is not clear enough.

The only difference between two kinds of sausages tested is the addition of sodium metabisulphite in the first product type which is completely absent in the second product. Although the residual content of sodium metabisulphite in the day of testing is well below the MRL (10 mg/kg). Presence of sodium metabisulphite acting as decolorant of azo dye complex is the cause of low nitrite level measured in the first product compared to the second ones even if the same initial sodium nitrite amount is used in both products.

Literature says that our intake of nitrate is from three main sources: vegetables, drinking water and food additives. (Merino et al, 2017). Among them, vegetables are generally the major source (75–91%), yet in some areas drinking water can account for the major contribution (European Commission Scientific Committee for Food 1997). This fact made us trying to explain why this nitrate content in both products when no nitrate is added. We checked all the other additives technical data sheet and the quality certificates of spices used in order to explain this fluctuating inconsistent nitrate presence. None of them could justify this presence.

After this check up we considered that this nitrate level may be the result of factors other than nitrate in spices e.g. contaminated water, but even the analysis of processing water resulted in a negligible amount at 6 mg/l nitrate expressed as KNO<sub>3</sub>.

Water used in establishment	NaNO <sub>2</sub> mg/kg	KNO <sub>3</sub> mg/kg
Process water	0	6

**Table 2:** Results of nitrite/nitrate in processing water

We already know that the main mechanism of nitro compound in meat products is the reduction of nitrate in nitrite serving as a nitrite reservoir for long term storage of products. But literature reports that nitrate is found in cured meat either through being used as a food additive or formed from conversion of initially added nitrite. If this is the case, the residual nitrate content is high enough to be derived from oxidation mechanism of initial nitrite amount. Summing up the contribution of all the potential exogenous sources such as water, other food additives, some nitrate present in nitrite salt used for meat curing, raw meat etc, it results that the major contribution of nitrate may be the biochemical conversion of nitrite to nitrate which leads to some nitrite loss.

**CONCLUSIONS**

There is a potential public health risk with elevated nitrate levels in food because of possible nitrosamine formation, as well as methaemoglobin condition specially in infants. Although in the light of increasing evidence, the reduction of nitrate to nitrite and the subsequent formation of biologically active nitrogen oxides could be beneficial. Even if these new studies are discovering interesting aspects of the biological chemistry of the nitrogen cycle, the presence of nitrate in food products, meat processed products in particular is of utmost importance, even though that meat products is the minor contributor of nitrate intake from consumers. Other factors may be of interest to be in control such as environment pollution which may be a potential contributor of nitrate intake from meat products ingredients, but however chemical biological mechanism of initial nitrite conversion could be major responsible for this exogenous spontaneous nitrate intake from consumers. Factors influencing to initial added nitrite to nitrate conversion should be further investigated as well as the correlation between sodium metabisulphite addition to nitrite/nitrate conversion in specific pH ranges.

**REFERENCES**

- Analysis of Nitrite and Nitrate in Foodstuffs  
Method development, occurrence, regulation, metrological aspects and exposure, D. Leonardo Merino, 2017  
Sulfites in meat: Occurrence, activity, toxicity, regulation, and detection. A comprehensive review, Teresa D'Amore Aurelia Di Taranto et al, 2020  
Effect of nitrite and nitrates on the microbiological safety of meat products, EFSA 2003, 2008