COMPARATIVE STUDY FOR DETERMINATION OF NITRATE IN MEAT PRODUCTS

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Abstract:

The aim of the present study is to offer alternatives regarding the determinations of nitrate from meat products. In this respect, the potentiometric determination of nitrate from these products was experimented, using a nitrate ion-selective liquid membrane electrode (NO_3-ISME). This method was compared with classical methods such as spectrophotometry and HPLC as regarding precision and determination speed. Based on the obtained results, we can conclude that for the meat products having a nitrate content situated in the linearity domain, from the point of view of electrode's Nernstian response, the potentiometric method is more accurate, less prodigious and faster than classical methods.

Keywords:

nitrate, potentiometry, NO_3-ISME, meat product

1. INTRODUCTION

Nitrate represents one of the most poisoning vectors from food, where it can be found because of an indirect way, as a consequence of nitrogen fertilization of plants, or by direct way, as a result of using it as additive in meat food products. As nitrite, nitrate poisoning can cause discoloration of the blood, due to the presence of methemoglobin. Nitrates in blood can also cause blood vessels to dilate and are responsible for peripheral circulatory failure. Other physical signs of nitrate poisoning include difficult and rapid breathing, muscle tremors, low tolerance to exercise, incoordination, diarrhea, frequent urination, which is why its use was severely restricted in many countries.

The numerous analytical applications of ISME, as well as the fact that potentiometric method based on such electrodes is very fast and accurate, encouraged us to experiment the use of the NO_3-ISME for the determination of nitrate ion in various food products.

2. EXPERIMENTAL

Equipment:

- HITACHI U 1100 spectrophotometer ;
- Merck-Hitachi L 6200A HPLC system ;
- pH/mV digital Hanna Instruments HI 8817;
- NO_3-ISME provided by Senzorom Cluj;
- Double junction reference electrode RA (0.3 M, Na_2SO_4 in the 2nd salt bridge) provided by Senzorom Cluj.

Reagents:

- 10^{-1}M and $5 \cdot 10^{-2}$ M, $NaNO_3$ stock solutions (with constant ionic force J = 0.1), prepared from $NaNO_3$ p.a., dried for 2 hours at 105°C;
- Solution for making dilution: 0.033 M, Na_2SO_4 solution ;
- Nitrate etalon solutions, prepared from stock solutions by diluting, with concentrations between the range 10^{-2} M÷$2.5 \cdot 10^{-4}$ M.

Extract preparation: 10 g of homogeneous sample, weightened with 0.001 g precision, is quantitatively passed through a calibrated 200 mL volumetric flask, by adding of 100 mL water. The content of the flask is warmed up to 60-70°C on a water bath, for 30 minutes, under vigorous stirring. The sample is cooled to room temperature, and than 2 mL Carrez I reagent and 2 mL Carrez II reagent are added one by one (for deproteinization), the solution being well stirred after each adding. The sample is leaving to repose for 30 minutes and than is filled with water to mark. The content of the volumetric flask is well homogenized and than is filtered [1].

The extract obtained by this procedure is used for the determination of nitrate from meat products by all three above mentioned methods.

3. RESULTS AND DISCUSSIONS

The potentiometric determination of nitrate ion from meat with NO_3-ISME was done by multiple standard addition, using 25 mL extract.

In case of potentiometric determination of nitrate, the main interference is given by chloride anion. In order to eliminate this interference, a 0.16 M, Ag_2SO_4 solution in ammoniac was used. The interference of hydrogen-carbonate is avoided by changing the pH of the solution to the value of 3.5 (with concentrated sulfuric acid solution). Other disturbing agents are nitrite and carboxyl anions, their action are removed with the help of a solution made up from: aluminum sulfate, sulfanilic acid and boric acid, having the pH=3.5 (fixed with a 0.1 N natrium hydricum solution) [2].

The calibration curve for the electrode and the plots resulted from experimental data by multiple standard addition [3, 4] is presented in figures 1 and 2.

The spectrophotometric determination of nitrate from meat products with brucine was done using 10 mL extract, prepared as mentioned above. The absorbance was measured at 410 nm comparatively with control [5, 6, 7]. The nitrate ion concentration from meat products was established by the help of a calibration curve plotted in the same experimental conditions (figure 3).

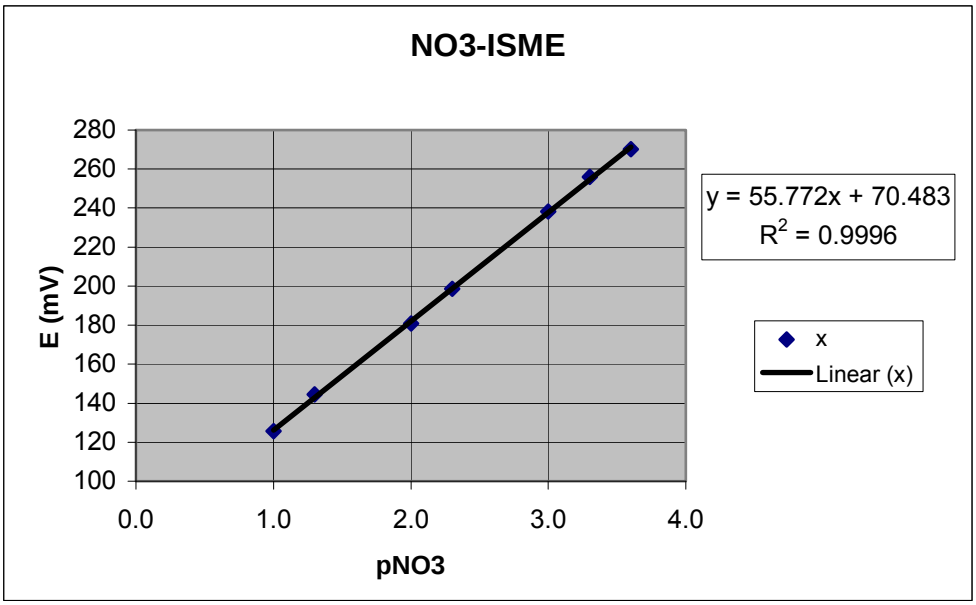


Fig. 1. *The transfer function of NO_3-ISME*

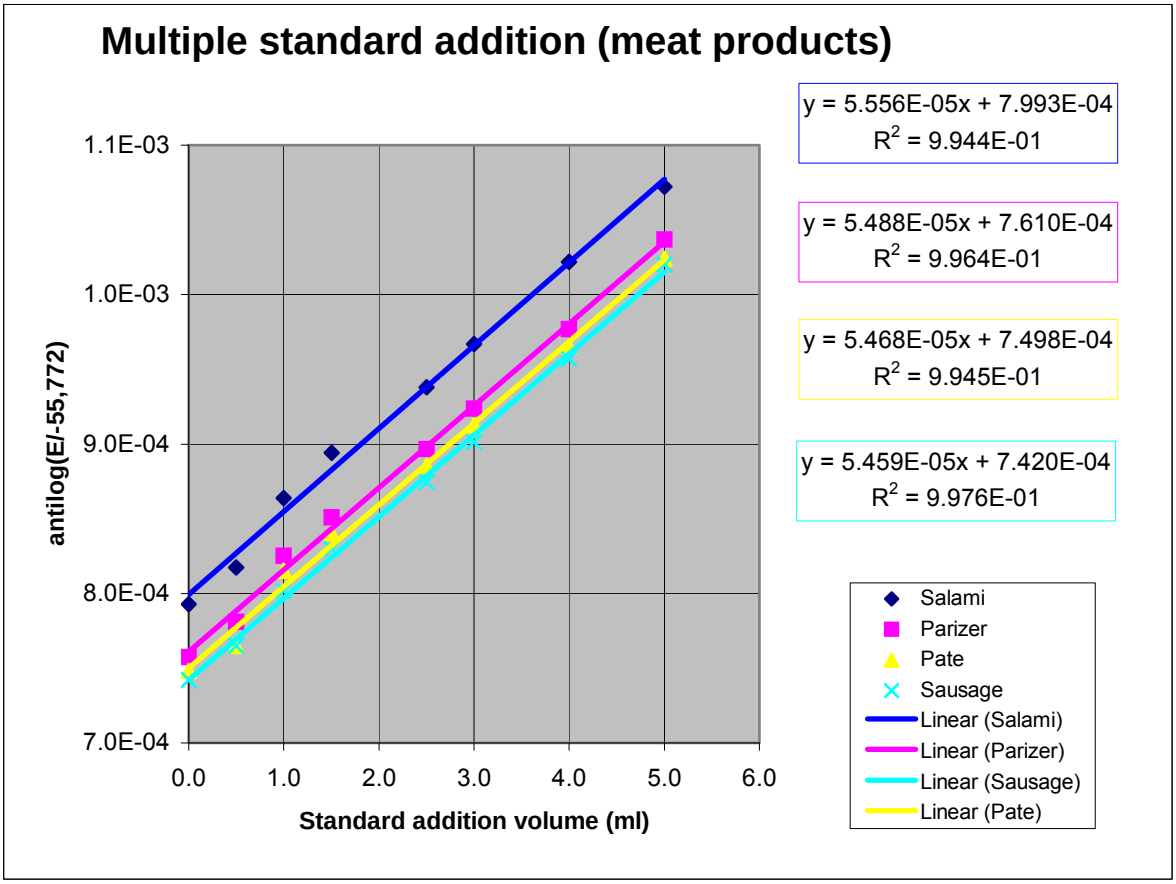


Fig. 2. *The determination of nitrate ion from meat products by means of multiple standard additions*

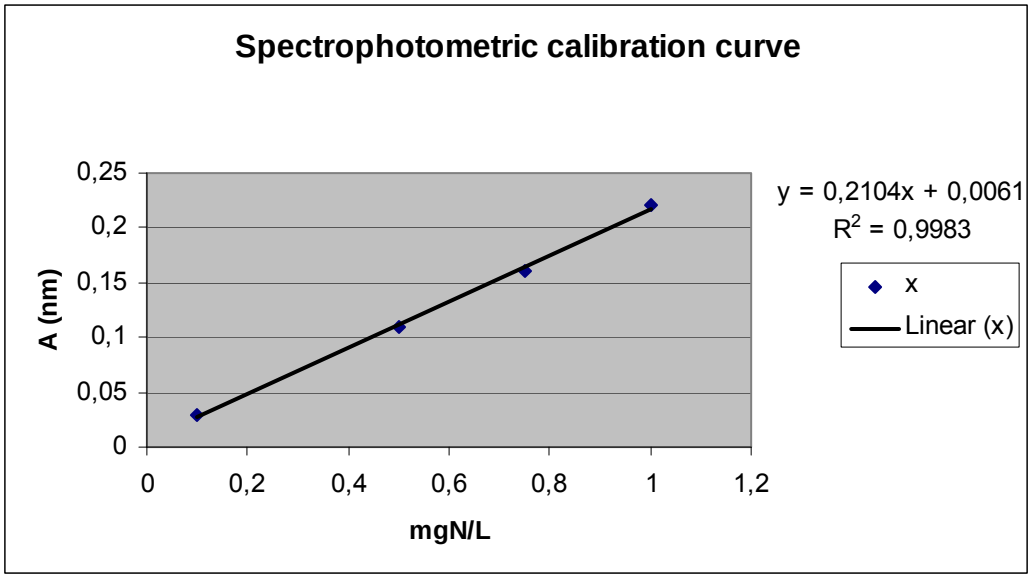
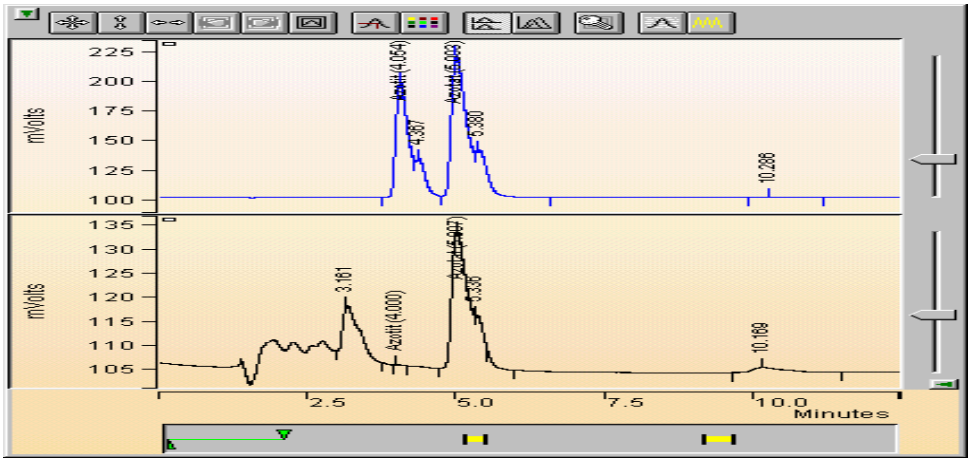
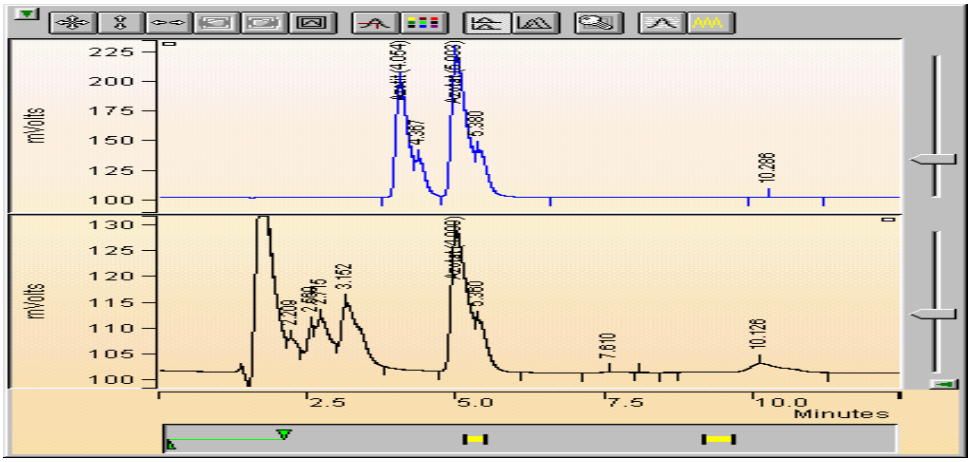


Fig. 3. *Spectrophotometric curve for the determination of Nitrate ion with brucine*

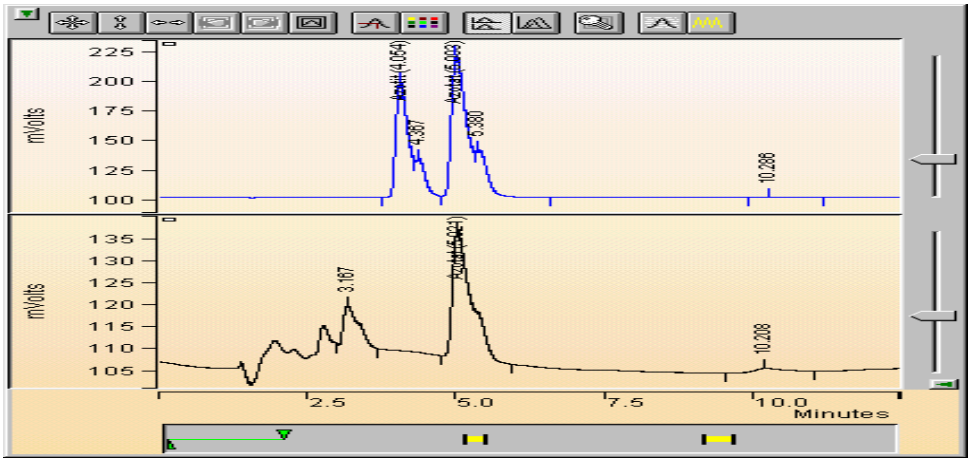
HPLC method for chromatographic determination of nitrate from meat, based on 50µL aqueous samples, and the results are illustrated in figure 4:



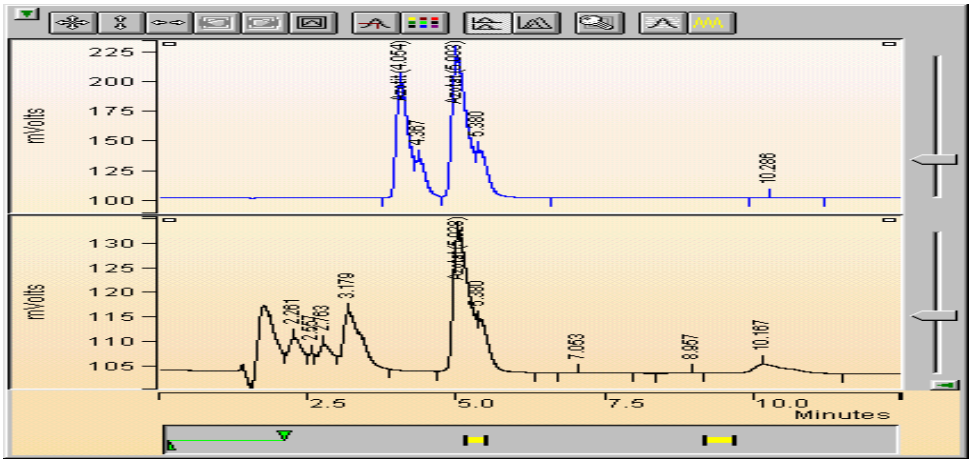
a)



b)



c)



d)

Fig. 4. *Up panel – mixture KNO_2 (252mg/L) / KNO_3 (249.5mg/L);*
Down panel – extract sample from:
a) sausage; b) parizer; c) pate; d) salami, 10 times diluted.
Column: SAX Nucleosil 100 SB, 150x4 mm, Φ = 5 μm. Mobile phase: 6.25% MeOH, 93.75% Phosphate buffer 45 mM (KH_2PO_4/K_2HPO_4), pH 6. Detection UV 210 nm, flow of mobile phase: 1 ml/min.

Comparative results at the determination of nitrate ion (mg NO /kg meat) by all the three methods: potentiometric, spectrophotometric and chromatographic, are presented in table 1:

Table 1. Nitrate content in different meat products

Meat product	Potentiometric	Spectrophotometric	HPLC
Salami	713.6 ± 3.1	713.5 ± 9.8	716.4
Pate	680.2 ± 2.3	681.3 ± 9.1	682.9
Parizer	687.8 ± 2.5	689.7 ± 9.4	686.8
Sausage	674.2 ± 2.1	668.6 ± 8.7	675.6

4. CONCLUSION

From the obtained data we can state that all the three methods lead to similar results.

The proposed method for the determination of nitrate from meat products using an NO_3-ISME represents an advantageous method due to its speed, the low cost of required equipment and also to its high precision.

The spectrophotometric determination method presents the drawbacks of a low reproductiveness, the need of higher cost reagents as well as a more laborious work. Besides, the reagent we used, brucine, is highly toxic.

The results obtained by HPLC are similar to those recorded by potentiometric and spectrophotometric methods and we have to mention that this method is easy to do and a small volume of the sample is needed, but the cost of the apparatus is too high.

5. REFERENCES

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