The Determination of Nitrite Content in Market Sausages

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Abstract. As a common food additive, nitrite exists widely in cooked meat foods such as sausages. This paper establishes a simple, sensitive and more accurate spectrophotometric method to measure the content of nitrite in sausage foods. After earlier period of treating the samples, the purple-red products formed with naphthyl ethylenediamine hydrochloride under weak acid conditions have the maximum absorption at 540nm. The content of sodium nitrite has a good linear relationship in the range of 0~0.40μg/mL by experimental analysis. The linear equation is A=0.6496C+0.0029. The correlation coefficient R^2=0.9969. The recoveries were in the range from 100.1% to 104.7%. The results of detecting the samples show that the nitrite content of the samples is between 2.8920 and 10.9498 mg/kg, which corresponds to the national limited standard.

1. Introduction

As nitrite reacts with protein to form carcinogenic compound nitrosamine, which increases the risk of cancer for people who like meat food [1] [2], so the amount of nitrite added in food has attracted great attention of researchers. At present, there are many methods for the determination of nitrite in food at home and abroad, which can be divided into two categories: spectroscopic method and chromatographic method [3] [4]. In addition to spectral method and chromatography, there are also electrochemical method and rapid detection method. In this study, visible spectrophotometry [5] was used to determine the content of nitrite in sausage. The operation method is simple and accurate, which can provide scientific basis for relevant food regulatory authorities.

2. Experimental

2.1. Experimental determination method

2.1.1. Determination of maximum absorption wavelength. Use a 1mL pipette to accurately transfer 0.40 mL to 25 mL of 10.0 μg/mL sodium nitrite standard solution into a colorimetric tube with stopper, add 2.00 mL of p-aminobenzenesulfonic acid, and mix well. After 4 minutes, add 1.00 mL naphthalene ethylenediamine hydrochloride solution into the solution, add distilled water to constant volume to the scale line, shake well, and let stand for 15 minutes. On the spectrophotometer, reagent blank was used as reference. Take the solution to be tested in the wavelength range of 500-610 nm, scan once every 10 nm, and record the absorbance value. The curve was drawn with abscissa as wavelength and ordinate as absorbance. The maximum absorption wavelength of azo compounds is 540 nm (Figure 1).

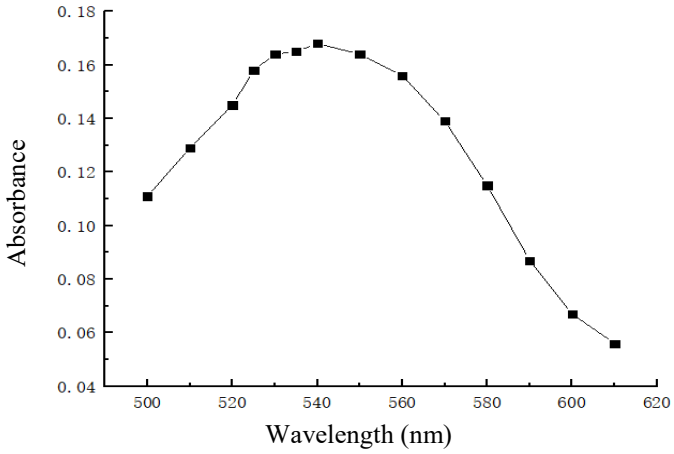

Figure 1. Absorption spectra of azo compounds

2.1.2. Drawing of standard working curve. Pipette 0.00 mL, 0.20 mL, 0.40 mL, 0.60 mL, 0.80 mLl and 1.00 mL of 10.0 μg/mL sodium nitrite standard solution were accurately transferred into six 25 mL colorimetric tubes, Add 2.00 mL of p-aminobenzenesulfonic acid in sequence and mix well, After standing for 4minutes, add 1.00 mL naphthalene ethylenediamine hydrochloride solution in turn, Add distilled water to volume to scale, mix well, stand and wait for 15 minutes. Taking reagent blank as reference, the absorbance was measured at 540 nm, the concentration of sodium nitrite was taken as abscissa, the measured absorbance value was as ordinate, the standard working curve was drawn, and the regression equation was calculated(Figure 2).

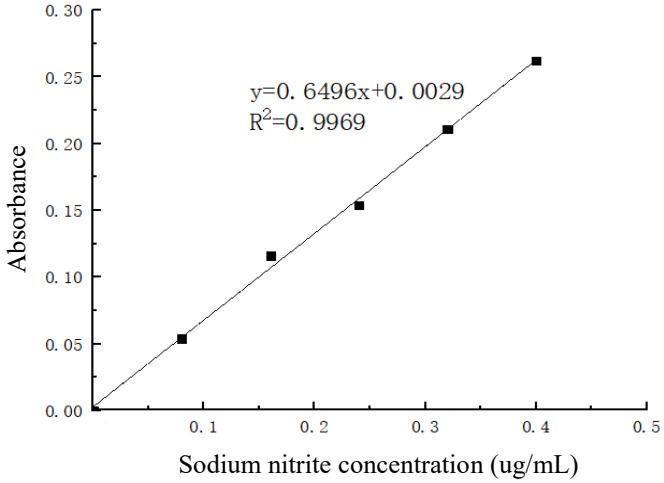

Figure 2. Standard curve of sodium nitrite

2.2. Optimization of experimental conditions

2.2.1. Effect of p-aminobenzenesulfonic acid on absorbance. Taking 0.40 mL sodium nitrite standard solution as an example, the sodium nitrite standard solution and 1.00 mL naphthalene ethylenediamine hydrochloride were added into seven 25mL colorimetric tubes in order.The dosage of p-aminobenzenesulfonic acid was different, which is 0.50 mL, 1.00 mL, 1.50 mL, 2.00 mL, 2.50 mL, 3.00 mL and 3.50 mL respectively. Mix them at constant volume and let them stand for 15minutes. The standard solution without sodium nitrite is used as blank control for determination. The results showed that the maximum absorbance value was obtained when p-aminobenzenesulfonic acid was added at 2.00 mL, which was the best addition value.

2.2.2. Effect of adding amount of Naphthylethylenediamine hydrochloride on absorbance. Taking 0.40 mL of sodium nitrite standard solution as an example, sodium nitrite standard solution and 2.00 mL p-aminobenzenesulfonic acid were added to seven 25 mL colorimetric tubes in turn, Add 0.60 mL of hydrochloric acid, 0.60 mL of naphthalene and 1.0 mL of ethylamine to the constant volume, After standing for 15minutes, the standard solution without sodium nitrite was used as blank control, and the absorbance was measured at 540 nm under the experimental conditions. The results show that the addition of naphthalene ethylenediamine hydrochloride has obvious influence on the absorbance value. With the increase of the content of naphthalene ethylenediamine hydrochloride, the absorbance first increased and then decreased. The maximum absorbance value was obtained when the dosage of naphthalene ethylenediamine hydrochloride was 1.00 mL.

2.2.3. Effect of color developing time on absorbance. Accurately transfer 0.40mL to 25mL of sodium nitrite standard solution into a colorimetric tube with stopper, add reagent to constant volume and mix well. The standard solution without sodium nitrite was used as blank reference, Under the experimental conditions, the absorbance values were determined at 540 nm after 5min, 10min, 15min, 20min, 25min, 30min, 35min and 40min. The results showed that the color development of developer was affected by time, and the change range was obvious before color development, and the absorbance value reached the maximum after 15minutes. After 15minutes, the change range of absorbance became smaller and no longer changed with time. Therefore, the best color developing time is 15minutes.

3. Sample determination

3.1. Optimization of experimental conditions

Five kinds of sausage or ham sausage with different flavors and brands, such as Shuanghui and Jinluo, were purchased from school supermarket and fortune spring shopping center. Five kinds of samples crushed by food processor were weighed and put into five 50mL beakers, and 6.00mL saturated borax solution was added into each beaker, and stirred evenly with glass rod.Add 80.00 mL distilled water at 70℃ and wash the sample in the beaker into a 250mL conical flask. Heat in a boiling water bath for 15minutes and remove. While rotating the conical flask, add 5.00 mL potassium ferrocyanide solution, shake the conical flask to mix evenly, and then add 5.00 mL zinc acetate solution to precipitate the protein in the sample.Then transfer it to a 100mL volumetric flask for constant volume, shake well, and let stand for 30minutes to remove the upper lipid in the liquid. After filtration, 70.00 mL of filtrate was obtained, 30.00 mL of initial filtrate was discarded, and the remaining filtrate was collected for standby.Then accurately transfer 20.00 mL of each of the five sample treatment solutions into five 25mL colorimetric tubes with stopper, add 2.00 mL of p-aminobenzenesulfonic acid to them, and mix them evenly. After waiting for 4minutes, add 1.00 mL naphthalene ethylenediamine hydrochloride solution into the colorimetric tube, add distilled water to the scale, mix evenly, and let stand for 15minutes. Use 1 cm colorimetric dish (moisten with the solution to be tested before determination), take reagent blank as reference, measure the absorbance at 540 nm, measure three times in parallel, take the average value, replace the measured absorbance value into the regression equation, and calculate the nitrite in the sample The content of sodium acid.

3.2. Determination results of samples

Table 1. Determination of sodium nitrite in seven samples

Sample	Quality/g	A_1	A_2	A_3	Average A	RSD/%	Sodium nitrite content mg/Kg
Sample1	15.8887	0. 552	0. 551	0. 552	0. 552	0.0013	8.3126
Sample2	15.6179	0. 614	0. 615	0. 614	0.614	0.0012	9.4116
Sample3	17.3080	0. 210	0. 212	0.211	0. 211	0.0047	2.8920
Sample4	16.6750	0. 761	0. 762	0.762	0. 762	0.00065	10.9498
Sample5	15.9556	0. 482	0. 483	0. 482	0. 482	0.0015	7.2225

The experimental data show that the content of nitrite in 7 kinds of samples are in line with the national standard, so they can be eaten safely.

3.3. The recovery experiment of sodium nitrite was carried out by adding standard

Transfer 0.80 mL, 1.00 mL, 1.20 mL, 1.40 mL and 10.0 μg/mL sodium nitrite standard solution into four 25mL colorimetric tubes respectively, add reagent to them according to the method for determination of sodium nitrite in the sample, add distilled water to constant volume, mix well, and measure the absorbance value. The recoveries of four samples with different concentrations are between 100.1% and 104.7%, which indicates that the method has good recovery effect and meets the requirements of analysis and determination.

4. Conclusion

In this paper, the absorbance of azo products was determined by visible spectrophotometer with naphthalene ethylenediamine hydrochloride as chromogenic agent. Draw the standard curve of sodium nitrite, and then calculate the regression equation according to the chart and data. After the sausage sample is pretreated, the absorbance is measured, and then the content of sodium nitrite in the sample can be calculated according to the linear equation. According to the hygienic standard for the use of food additives in China, the upper limit of nitrite used in cooked meat products is 0.15g/kg. The experimental data show that the nitrite content in the seven samples is between 2.8920-10.9498mg/kg, which meets the national standard and can be eaten safely. In addition, the determination conditions were optimized, and the recoveries were between 100.1%-104.7%, with good accuracy.

Acknowledgments

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