The Effect of Boiling Time on the Concentration of Nitrite in Sausages

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Abstract

Background : Nitrite is often used as a preservative as well as curing to brighten foods made from raw meat such as sausages. Excessive use of nitrite can cause symptomatic methemoglobin, tumors to cancer in humans. One method that can be used to reduce nitrate levels in sausages is by boiling.

Aim : The study aimed to determine the effect of boiling time on the nitrite content of sausages.

Material and Method : This type of research is an experiment with a one group

pretest-posttest design. The research material is sausages obtained from distributors, Banjarbaru. The research material treatment was carried out by boiling sausages with distilled water for 5, 10, 15, 20, and 25 minutes at 80-90^{o}C.

Results : There was a decrease in the level of nitrite in sausage after boiling, which is consecutive at 1.44 ppm (5 min); 1.17 ppm (10 minutes); 0.98 ppm (15 minutes); 0.83 ppm (20 minutes) and 0.7 ppm (25 min). The linear regression test found that R square was 0.574 or 57.4%.

Conclusion : Meat sausage boiling process can lower nitrite levels in sausages, the longer the boiling process is increasing the amount of nitrite being eliminated from the sausage with the highest number of boiling for 25 minutes there is a decrease of 70%.

Keywords*: Sausage, nitrite level, boiling time*

Introduction

Meat is one source of animal protein that is needed by humans protein functions for cell growth, replacement of damaged cells, and as fuel in the human body. In addition to protein, meat has other components such as minerals, carbohydrates, and fats that cause meat easily damaged, especially by microorganisms such as fungi and bacteria. To maintain the freshness of meat-based foods such as sausages, nitrite preservatives often added by producers. The nitrite preservation mechanism is using nitrite binding to the sulfhydryl group to form salts which are challenging to metabolize by microbes in anaerobic conditions[1,2].

Nitrite is one of the preservatives that is allowed to use based on Republic of Indonesia Minister of Health Regulation No. 1168/Menkes/Per/X/1999 concerning Food Additives, with a limitation of the maximum use of nitrite preservatives in processed meat products of 125 mg/Kg. Nitrite is widely used in meat because in addition to preventing microbial growth it can also function as curing or improving the color of meat. But the risk of nitrite use limits its use. Nitrite in the beef will react with oxygen to form nitroxide which will then respond with pigments in the flesh to build nitrosomyoglobin, which gives a bright color to the flesh. Furthermore, in the body of the nitrite can also bind to amino or amide in the body to form derivatives of nitrosamines, which are toxic and carcinogens[3].

As the food and beverage industry develops, more food products from meat are produced, sold and consumed in forms that are more durable, attractive and more practical than fresh products such as bread, meatballs, nuggets, sausages and corned beef. There are still many food products currently circulating that do not

meet the hygiene requirements and food safety quality standards. In terms of microbiological safety, some food products are still found to be fungal[4] contamination, Salmonella[5], Staphylococcus[6,7], Bacillus[8,9]. In terms of chemical safety of many foods and beverages in circulation that still use food additives that not permitted for food and the use of doses that exceed the required threshold for permitted food additives such as the use of nitrite.

The case of nitrite poisoning in sausages had occurred in 2008 at Sukosewu 1 Gandusari Blitar elementary school. The students poisoned after eating sausages sold at the school. Nur's HH reported that five sausage brands examined all positively contained nitrites and there was one brand of sausage with nitrite levels of 208.19 mg/Kg which exceeded the quality standard according to Permenkes No.1168/Menkes/Per/1999 which is equal to 125 mg/Kg[10].

The use of nitrite compounds in processed meat products such as sausages seems to have become a necessity of the community because nitrite compounds in meat besides acting as preservatives also provide a distinctive color of fresh red meat. However, the use of nitrites in food must remain vigilant because if consumed excessively and repeatedly it can adversely affect health either directly or indirectly, among others, can result in methemoglobinemia, teratogenic, tumors to cancer through a reaction between secondary or smear amines contained in the body[11].

One way that can be done to minimize nitrite content in doses is by boiling before consumption — this theory supported by the results of research conducted by Li[12]. The results of Li's research proved to be able to reduce nitrite levels in sausages by 19.47% after boiling for 60 minutes at 80ºC. Study of the effect of boiling time on nitrite levels in sausages by spectrophotometry using the Griess method.

Methods and Materials

This type and design of the study were experiments with the study design one group pretest-posttest. The research material is unbranded sausage from processed beef, which is known to have positive nitrite content after a preliminary qualitative test was carried out using the Griess method. The research instruments consisted of UV-Vis spectrophotometer. The reagent used was Natrium nitrite, Sulfanilic acid, Naphthylenediamine, 30% Acetic acid, Glacial acetic acid, quads.

Sausages treated with five sausages each cooked with 500 ml of water with variations time of 5 minutes, 10 minutes, 15 minutes, 20 minutes, and 25 minutes at a temperature of 80-90ºC. After that, the sausages are taken to check each nitrite level with four repetitions. Analysis of nitrite examination carried out qualitatively and quantitatively. A qualitative study using Griess reagent based on the formation of the color of azo substances. Quantitative analysis using spectrophotometric methods[13].

The sausage weighed and then it is heated as much as 5 grams in a 50 ml glass beaker, add 50 ml of hot distilled water, stirring and strain. The filtrate was taken as much as 25.0 ml and put in a 50 ml volumetric flask, diluted with distilled water until the boundary mark. Add 4.0 ml of Griess reagent. Leave it for 15 minutes, put it in the cuvette, and read the absorbance on the spectrophotometer with a wavelength of 548 nm. Nitrite levels at doses are calculated based on the line equation $Y = bX + a$. It repeated for variations time of sausage 10, 15, 20, and 25 minutes.

The making of the standard sodium nitrite curve solution is each with a concentration of 0.2; 0.6; 1.0; 1.4; 1.8 and 2.2 ug/ml into 50.0 ml volumetric flasks then advert with distilled water. Then 10.0 ml of each solution was taken, and then 2.0 ml of Gress reagent was added to each 50 ml volumetric flask and the boundary mark. The answer was left operating time for 30 minutes then read the absorbance value using a UV-Vis spectrophotometer (what brand) at a maximum wavelength of 548 nm. The absorbance result data is made a standard curve so that the line equation $y = bX + a$ obtained. This line equation is used to determine sodium nitrite levels in sausages. 2). Determination of nitrite levels in doses, namely as much as 5 grams of mashed sample put into a glass beaker plus 50 ml of distillate water temperature 80oC, strain. The 25.0 ml filtrate was put into a 50.0 ml volumetric flask and then diluted with distilled water to the boundary mark, plus 4.0 ml Griess reagent, homogenized, the solution left for 30 minutes and measured the absorbance by a spectrophotometer at a wavelength of 548 nm.

Result and Discussion

Data on the results of the examination of nitrite levels in sausages that have boiled for 5, 10, 15, 20, and 25 minutes with four repetitions can be seen in table 1 and figure 1.

Table 1. Nitrite Level in Sausage

Boiling Time	Nitrit (ppm)				Average (ppm)	Reduction in nitrite levels (%)
	I	II	III	IV		
0	2.64	2.64	2.64	2.64	2.64	0
5	1.60	1.33	1.11	1.72	1.44	40
10	1.15	1.06	1.01	1.46	1.17	55
15	1.05	0.99	0.69	1.22	0.98	62
20	0.93	0.77	0.47	1.18	0.83	68
25	0.77	0.71	0.38	0.96	0.70	70

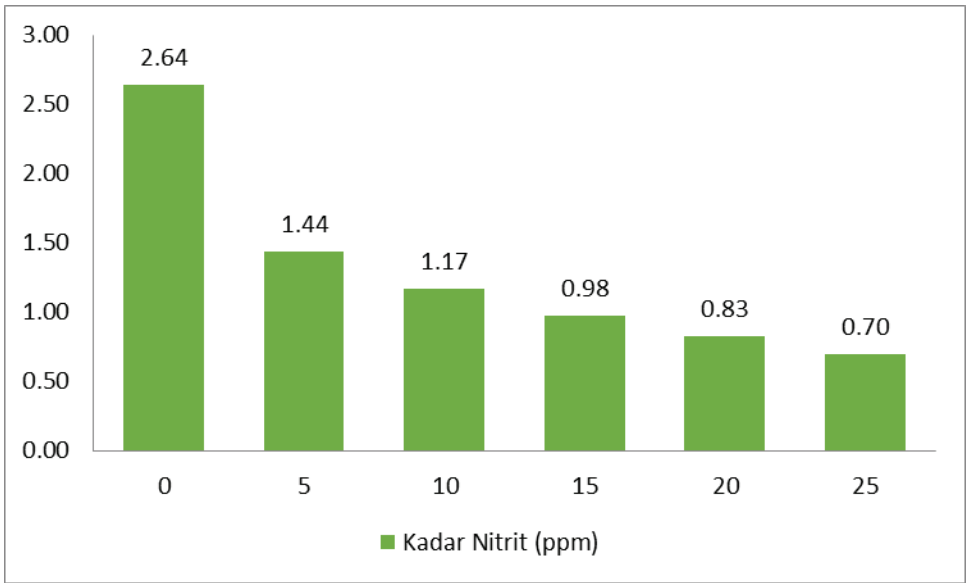

Figure 1. Nitrit level reduction in sausages after boiling with different temperature variations

Based on Figure 1, it is known that the longer the sausage boiled, the less the amount or level of nitrate in sausages. This result is somewhat different from the research conducted by Li6 on sausages, namely the reduction of nitrite in sausages only by 19.47% after boiling for 60 minutes at 80ºC.

The difference in nitrite levels in each treatment in this study caused by the difference in the length of sausage boiling. The longer the boiling time, the more nitrite is eliminated from the sausage. The highest percentage of nitrite reduction in sausages occurred at 25 minutes of boiling time, which is 70% and the lowest in sausages with a boiling time of 5 minutes, which is equal to 40%.

This study using the Griess method spectrophotometer. The principle is the Griess method based on the diazotization reaction, which is between the nitrite ion in the sample and sulfanyl acid in an acidic atmosphere to form benzene diazonium ions. Furthermore, benzene diazonium ions coupled with N-1-Naphthylen-diammonium dihydrochloride (NEDA) which will create a purple-red azo compound which can be measured absorbance at the maximum wavelength obtained in this study is 548 nm[13,14].

the use of excess nitrite is carcinogenic and mutagenic because nitrite in the body can bind to secondary and tertiary amines to form derivatives of nitrosamines, especially at low pH and salt levels. Carcinogenic and mutagenic properties of nitrosamines are thought to be the cause of necrosis, which is the death of some cell tissues of living creatures as an early stage of cancer attack[14]. One of the advantages of nitrosamines is its capacity to cause tumors in several organs, including liver, kidney, stomach, bladder, esophagus, and SSP[10]. Other than that, nitrite can also bind to Fe (II) contained in blood hemoglobin, which then forms methemoglobin which cannot carry oxygen, which can cause the body to experience oxygen deficiency. If this is allowed to continue, it can have fatal consequences primarily if it occurs in infants and pregnant women with symptoms of pallor, cyanosis, vomiting, shortness of breath to shock[15].

Taking into account the negative effects of nitrites in processed meat foods such as sausages, several ways can be done to reduce nitrite content before consumption, one of which is to boil sausages using water for a certain amount of time before consumption. In this study, boiling sausages for 5-25 minutes with a temperature of 80-90OC was shown to show different variations in nitrite reduction according to the length of boiling (see table 1). In another journal, it also reported that the use of nitrite in sausages eliminate by adding various combinations of ingredients such as celery, carrots, broccoli and ingredients that contain vitamins C and E during boiling sausages[15,16]. In addition to boiling methods, the use of several types of natural dyes can also be an alternative as a red coloring in meat without causing health problems, such as angkak[17], bits (Beta vulgaris)[3], the skin of red dragon fruit[18]. Cochineal 0.015% can also be used as a substitute for nitrite in meat because it can develop the color of meat and is quite stable to light and pH[19]. Some spice plant extracts such as cloves, lemons, licoric are also known to have antibotulinin activity in processed meat[20]. Celery powder with a concentration of 10% can protect sausages from deterioration during storage as a substitute for nitrite[21]. With several alternatives that can be used to replace nitrite as curing as a preservative in sausage meat it is hoped that food producers who still use nitrite could replace it with natural, safer and friendlier materials for the body.

Measurement of sample levels in this study using the calibration curve method, namely the standard curve of the standard solution of sodium nitrite. The purpose of making this standard curve is to get more accurate results because it uses more than one concentration of different standard solutions, which commonly referred to as standard series solutions. In this study using 6 standard series solutions each 0.2; 0.5; 1.0; 1.4; 1.8; and 2.2 ug / mL or (ppm). Based on the results of measurements of the absorbance of standard solutions obtained by absorbance in a row 0.125; 0.186; 0.217; 0.312; 0.384 and 0.481. From the results of the calibration curve, the linear regression equation y = 0.1763x + 0.0725 with R2 is 0.9788. Furthermore, this line equation is used to determine the nitrite level in the sausage sample by entering the absorbance of the measured sample. The results of the calculation of nitrite levels in sausages that have undergone boiling with various variations of boiling time can see in table1.

To find out whether there is an influence of the old boiling variable (independent variable) on nitrite levels (dependent variable) on sausages and how much influence the regression test linear. Previously, a regression test conducted before the normality test was carried out on the data to find out whether the data were normally distributed or not. From the test results, it knew that the significant value is higher than α (0.05) so that the data usually distributed. Furthermore, the ANOVA test was carried out to find out the differences between the data groups and obtained a significant value of 0,000 which means that it was higher than α (0.05) so that there were significant differences between the data groups. Based on the linear regression test, the R Square value is 0.574 or 57.4% so that it can say that the length of sausage boiling has an effect of 57.4% on nitrite levels in doses.

Conclusion

Based on the results of the study it can be concluded that the boiling process of meat sausages can reduce nitrite levels in sausages, the longer the boiling process increases the amount of nitrite eliminated from sausages with the highest number of boiling for 25 minutes a decrease of 70%.

Conflict of Interest: Nil

Source of Funding: Self

Ethical Clearance: Taken From Health Research Ethics Committee Politeknik Kesehatan Banjarmasin indonesia

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