

Microbiological safety of processed meat products formulated with low nitrite concentration — A review

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Abstract: Nitrite plays a major role in inhibiting the growth of foodborne pathogens, including *Clostridium botulinum* (*C. botulinum*) that causes botulism, a life-threatening disease. Nitrite serves as a color-fixing agent in processed meat products. However, *N*-nitroso compounds can be produced from nitrite, which are considered as carcinogens. Thus, consumers desire processed meat products that contain lower concentrations (below conventional concentrations of products) of nitrite or no nitrite at all, although the portion of nitrite intake by processed meat consumption in total nitrite intake is very low. However, lower nitrite levels might expose consumers to risk of botulism poisoning due to *C. botulinum* or illness caused by other foodborne pathogens. Hence, lower nitrite concentrations in combination with other factors such as low pH, high sodium chloride level, and others have been recommended to decrease the risk of food poisoning. In addition, natural compounds that can inhibit bacterial growth and function as color-fixing agents have been developed to replace nitrite in processed meat products. However, their antibotulinal effects have not been fully clarified. Therefore, to have processed meat products with lower nitrite concentrations, low pH, high sodium chloride concentration, and others should also be applied together. Before using natural compounds as replacement of nitrite, their antibotulinal activities should be examined.

Keywords: Processed Meat; Nitrite; Microbial Safety; Substitute

INTRODUCTION

Nitrite has been used in processed meat products to enhance color, flavor, storage, and antioxidant activity. It is also used to control foodborne pathogens and lipid rancidification [1,2]. Under anaerobic environmental conditions, nitrite can control *Clostridium botulinum* (*C. botulinum*) germination and the growth of various foodborne pathogens such as *Listeria monocytogenes*, *Clostridium perfringens*, *Achromobacter*, *Aerobacter*, *Escherichia*, *Flavobacterium*, and *Micrococcus* spp. [3]. However, nitrite can be converted into *N*-nitroso compounds known as carcinogens by responding to amines generated from products under acidic conditions, and such conversion can be accelerated by high cooking temperature [3-4]. Therefore, safety issues about nitrite have been raised [2]. Meat products with lower nitrite (below conventional concentration of products) concentrations have been produced [5]. However, Doyle and Glass [6], and Gwak et al [7] have suggested that there are problems related to microbial safety of processed meat products with no or low nitrite concentrations. Therefore, the objective of this short communication was to review food safety of processed meat products formulated with no or low nitrite concentrations.

INTAKE OF NITRITE THROUGH CONSUMPTION OF PROCESSED MEAT

The U.S. limits the ingoing level of sodium (or potassium) nitrite for curing. The maximum nitrite levels for comminuted, massaged or pumped, immersion cured, and dry cured meat and poultry products are 156, 200, 200, and 625 ppm, respectively [8]. Maximum sodium nitrite levels for immersion-cured bacon, pumped and/or massaged bacon, and dry cured bacon are set at 120 (or 148 ppm potassium nitrite), 120 (or 148 ppm potassium nitrite), and 200 (or 246 ppm potassium nitrite) ppm, respectively [8]. EU and European countries have also limited the ingoing level of sodium (or potassium) nitrite for processed meat products. The maximum ingoing level of nitrite for cured and canned meat products is 150 ppm in EU [9]. Codex Committee on Food Additives (CCFA) also has regulation for nitrite levels in heat-treated processed meat and processed comminuted meat. For cured ham and cooked cured pork shoulder, the maximum residual nitrite level is 80 ppm. Also, residual nitrite levels in luncheon meat, cooked cured chopped meat are limited to 80 ppm except corned beef whose maximum residual nitrite level is 30 ppm [10]. Korea and Japan mandate the residual nitrite concentration be <70 ppm for processed foods (such as meat products, meat extract processed products, edible beef tallow, and edible pork), up to 50 ppm for fish sausages, and 5 ppm for salted pollack roe and salmon roe [11,12]. However, these concentrations are not observed in processed meat products in markets because nitrite levels in products will decrease continuously due to storage temperature, acidity, heating temperature, storage time, and the presence of other food additives [3]. In the US, levels of residual nitrite and nitrate in cured meat products are 0.64 to 7.31 and 14.81 to 78.81 ppm, respectively [13]. In South Korea, average residual nitrite levels for ham, sausage, and bacon are 16.6, 4.6 ppm, and 15.8 ppm, respectively [14], which are much lower than concentrations listed in regulations. Archer [15] has suggested that ingested nitrites and nitrates are mostly from vegetables and saliva rather than from cured meat. Chung et al [16] have reported that lettuce and spinach have nitrate concentrations of 2,430 and 4,259 ppm, respectively. These are considerably higher than those in meat products reported by Keeton et al [13]. White [17] has found that 81.2% of nitrate

intake by Americans is from vegetables, which is higher than nitrate intake from cured meat products (14.7%). Hord et al [18] have also estimated that around 80% of dietary nitrates come from vegetables. Consumed nitrate is absorbed to the plasma through the proximal small intestine [19], and about 65% to 70% of nitrate in plasma is excreted by passive urinary secretion. There are two active secretion systems, colonic and salivary, and the latter is more important due to 25% of entered nitrate is recycled via salivary secretion [19]. One fifth of the recycled nitrate (ca. 5% of the total entered nitrate) is converted to nitrite by microorganisms in oral cavity [19,20]. The nitrite from salivary source account for about 93% of the total ingested nitrite even though it seems like small amount [21]. These results indicate that the portion of nitrite intake by consuming processed meat might be low among total nitrite intake.

MICROBIAL INHIBITION BY LOW NITRITE CONCENTRATIONS

Nitrite in processed meats can inhibit the growth of foodborne pathogens and food spoilage bacteria through various mechanisms, including oxygen uptake and oxidative phosphorylation interruption, formation of nitrous acid and NOs, and interruption of critical enzymes in bacterial metabolism such as aldolase [22]. Nitrite has been used to prevent and control *C. botulinum* growth and other pathogens in processed meat products (Table 1). When nitrite concentration is increased, inhibition for the growth and toxin production of *C. botulinum* is also increased [23]. Robinson et al [24] have examined the effect of nitrite on *C. botulinum* toxin production and reported that the addition of 100 ppm nitrite to pasteurized and cured meats can result in 59% probability of toxin production by *C. botulinum*, which was much higher than the 1% probability of toxin production by *C. botulinum* in the presence of 300 ppm nitrite during storage at 20°C. After 84-day of storage at 27°C, *C. botulinum* toxin has been detected in 7 of 10 bacon samples containing 60 ppm nitrite. However, *C. botulinum* toxin was detected in only 1 of 10 bacon samples containing 340 ppm nitrite [25]. Cui et al [11] have suggested that a significant reduction in nitrite concentrations might lead to increased risk of food poisoning caused by *C. botulinum*. A

Table 1. Various nitrite levels to control microbial growth

Object	Target bacteria or toxin	Nitrite levels for inhibition of microbial growth (ppm)	Reference
Pasteurized and cured meats	<i>C. botulinum</i> toxin	100, 300	[25]
Bacon	<i>C. botulinum</i> toxin	60, 340	[26]
Meat products	Spore-forming pathogens (e.g. <i>C. perfringens</i> and <i>B. cereus</i>)	100-200	[32]
Broth	<i>C. perfringens</i>	50, 100	[33]
Pork sausage	<i>E. faecalis</i>	200	[34]

lowered concentration of nitrite in meat products can result in overgrowth of food-spoilage bacteria (*Lactobacillus* spp., *Enterococcus* spp., and *Pseudomonas* spp.) and foodborne pathogens (*L. monocytogenes*, *Salmonella*, and *Staphylococcus aureus*) in processed meat products [5,26-30].

Nitrite is also effective against other spore-forming pathogens such as *C. perfringens* and *Bacillus cereus* because 100 to 200 ppm nitrite can prevent the outgrowth of germinated spores [31]. Myers et al [32] have investigated the effect of lower nitrite concentrations (0, 50, and 100 ppm) on *C. perfringens* outgrowth in broth system during thermal process and cooling cycle used for processed meat products. It has been found that both nitrite concentration and temperature have significant effects on *C. perfringens* populations and spore survival, suggesting that lower nitrite concentrations are associated with a risk of *C. perfringens* outgrowth [32]. Sameshima et al [33] have reported that the growth of *Enterococcus faecalis* at 10°C is twice slower in pork sausage containing 200 ppm nitrite under vacuum packaging compared to that in sausage containing no nitrite. These results suggest that lower nitrite concentrations will allow more toxin production of *C. botulinum* and more growth of foodborne pathogens in processed meat products. Therefore, the overgrowth of pathogens in meat products formulated with lower nitrite concentrations should be controlled with other combinations.

COMBINATIONS TO IMPROVE MICROBIOLOGICAL INHIBITION

Gunvig et al [34] have developed a model to predict the interface between growth and no growth of *C. botulinum* in meat products (bologna type sausage, chicken cold cuts, and pork cold cuts), containing various levels of sodium nitrite (0 to 150 ppm), sodium lactate (0 to 3%), and sodium chloride (1.2% to 2.4%) with different pH levels (5.4 to 6.4) at 4°C to 12°C. They found that *C. botulinum* did not grow in the presence of 72 to 150 ppm nitrite irrespective of pH, levels of sodium chloride, levels of sodium lactate, or temperature. However, in the presence of 60 ppm nitrite, growth of *C. botulinum* was increased at pH>6, higher storage temperatures, and lower concentrations of sodium chloride and sodium lactate. Sixty ppm nitrite was not sufficient to inhibit *C. botulinum* [34]. Thus, the use of other antimicrobial systems is recommended for products with such low levels of nitrite. Johnson [35] has reported that the minimum pH value for *C. botulinum* growth was 4.6 for Group I and 5.0 for Group II. The minimum sodium chloride concentration for inhibiting *C. botulinum* growth was 10% for Group I and 5% for Group II. Recently, Doyle and Glass [6] have assessed the effect of a combination of sodium chloride and sodium nitrite on *L. monocytogenes* growth, using ComBASE (www.combase.cc/) and found that the antilisterial effect of 100 ppm sodium nitrite plus 0.5% to

5.5% sodium chloride is increased with increasing sodium chloride concentration. Harrison et al [36] have reported that ground beef jerky with a nitrite and salt cure mix can decrease *E. coli* O157:H7 cell counts compared to meat without such cure mix during drying. However, *S. aureus* is resistant to 8% sodium chloride and up to 100 ppm nitrite [37]. In the presence of 100 ppm nitrite at pH 6.0, a 4-log increment of *L. monocytogenes* cell counts in the presence of 0.5% sodium chloride has been observed after storing at 37°C for 8 h or 5°C for 287 h. However, in the presence of 4.5% sodium chloride, storing at 37°C for 11 h or 5°C for 479 h is required to induce a 4-log increase of *L. monocytogenes* [38]. McClure et al [39] have reported that *L. monocytogenes* does not grow within 21 days at 20°C and pH 5.3 with addition of 50 ppm nitrite. Indeed, at pH 6.0 and 10°C, the inhibitory effect of nitrite is minimal after adding 400 ppm NaNO₂. Therefore, microbiological safety of processed meat products with lower nitrite levels can be improved by a combination of pH and sodium chloride concentration. Gwak et al [7] and Tompkin [40] have also suggested that increased sodium chloride levels can control the growth of harmful bacteria in processed meat products with lower nitrite concentrations because the antimicrobial effect of nitrite is influenced by sodium chloride concentration, pH, reductants, iron content, and so on.

MICROBIOLOGICAL SAFETY OF NATURAL SUBSTITUTES

Although some consumers prefer no or low nitrite in processed meat products [7], removing nitrite from meat products could be problematic because of its role in microbiological safety. Thus, the food industry has tried to replace synthesized nitrite with vegetables with high nitrite concentrations because consumers are more comfortable with natural sources. Hence, numerous studies have been conducted on natural sources of nitrite. Horsch [41] has compared the antilisterial effect of nitrite with celery juice which contains a high concentration of nitrite and found that the antilisterial effect of sodium nitrite is higher than that of celery juice at the same nitrite concentration. However, King et al [42] have suggested that equivalent nitrite concentrations from purified nitrite and celery juice powder have equivalent effectiveness on the growth of *C. perfringens*. Although these natural substances possess antibacterial and color-formation effects, and they could be regarded as nitrite substitutes, their fundamental ability to inhibit germination of *C. botulinum* spores should also be examined prior to their industrial use. Cui et al [11] have recently examined antibotulinal activities of 90 fresh herbs and spices, and found that nutmeg (0.05%), sage (0.02%), and clove (0.05) extracts possess antibotulinal activities in a model meat product without compromising its organoleptic properties.

CONCLUSION

The portion of nitrite intake through consuming processed meat in total nitrite intake is relatively low. However, consumers still want to have low nitrite concentration in processed meat products. Low or no nitrite concentration in processed meat products may increase the risk of illness caused by foodborne pathogens. Thus, other factors such as low pH, addition of sodium chloride, antimicrobials, and/or natural sources should be combined with nitrite to control foodborne pathogens, particularly for *C. botulinum* and *L. monocytogenes*. In addition, although some natural substances possess antimicrobial and color-formation effects, their antibotulinal activity must be examined before using them in processed meat products.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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