Use of Nitrite and Nitrite-Sparing Agents in Meats: A Review

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ABSTRACT

Nitrite plays a major role in the curing of meats. However, the potential problem of nitrosamine formation has been responsible for reduction or elimination of nitrate and nitrite in curing. Reduced amounts of nitrite in curing can provide less protection against botulism, and subsequent investigations have examined solutions to this situation. Residual nitrite has been reduced by limiting the ingoing nitrite and by introducing curing substances, i.e., phosphates, lactobacillus cultures, and phenolic smoke compounds, to lower the pH. Ascorbates, alpha-tocopherol and other blocking agents have been used with nitrite in an attempt to devise a curing system that can provide a safe product with the color and flavor associated with nitrite-cured products while retarding nitrosopyrrolidine formation. This review focuses on the efficacy of various curing methods using nitrites that have the goal of reducing formation of nitrosamines.

Meat has been cured for many centuries for preservation and development of flavor and color. Although the origin of the use of nitrates is not known, nitrates or nitrites in some form have been included in most cure formulations since curing has been practiced. Even though nitrites and nitrates were discovered as impurities in most salt previously used in curing, their use as additives has been approved and regulated by Federal agencies only since 1923.

As a result of early work by Polenske (39) and Haldane (18), technology was established which revealed that the nitrite found in cured meat and curing brine resulted from bacterial reduction of nitrate. During this century, it was also discovered that nitrite instead of nitrate is responsible for the characteristic cured meat color. During the early part of this century, the mechanisms of curing and the modification of pigments by cooking were studied by Hoagland (20). This knowledge was followed by an attempt of the U.S. Department of Agriculture to establish proper levels of nitrite for use in curing.

During the past 50 years curing progress has been related primarily to decreased curing time with consequently increased productivity. Since the early 1960s, acidulating agents have been used to decrease curing time, because of increased demand for cured meats (26). Curing time was further reduced during the 1960s by direct use of nitric oxide gas and chopping and mixing meat under vacuum.

During the latter part of the 1970s, most of the technological emphasis on curing has involved problem-solving. A major concern recently has been the possible formation of carcinogenic nitrosamines in meats cured with nitrite. Fiddler et al. (15), Lijinsky and Epstein (29) and Wolf and Wasserman (54) reported that nitrosamines produced as a result of interaction of secondary amines with sodium nitrite are carcinogenic in laboratory animals. Past reviews that address this problem have been published by Crosby and Sawyer (10), Sebranek (43) and Sofos et al. (47). Other research at the Massachusetts Institute of Technology (3) has suggested the possible carcinogenic activity of nitrite itself. The purpose of this review is not to provide another expansive review but to identify and discuss research that has dealt with curing compounds used with nitrite to reduce formation of N-nitrosamines and provide a safe, attractive, and flavorful product.

NITRITE AND NITROSAMINES

The potential problem of nitrosamine formation has been responsible for reduction or elimination of nitrate in most cure formulations and reduction of nitrite levels in many curing processes. This change in curing formulation serves as a potential method of reducing nitrosamine formation. The technological change has been responsible for almost complete elimination of nitrosamine formation in all cured meats except dry cured and pumped bacon (32).
NITRITES AND BOTULISM INHIBITION

Restricted use of nitrite in a cure formulation presents increased food safety risks. Ingram (23) suggested that 5-20 ppm of formulated nitrite would provide adequate color, but that more than 100 ppm of nitrite was needed to retard proliferation of Clostridium botulinum. Although researchers have not exactly determined the relative importance of formulated or residual nitrite for retarding growth of C. botulinum, it has been suggested that ascorbate or erythorbate can enhance the antibotulinal properties of nitrite in a synergistic manner to combat C. botulinum (51,52). Fox and Nicholas (16) and Mirvish and Shubik (31) have suggested that ascorbate and erythorbate reduce potential nitrosamine formation. Borenstein (6) reported that ascorbate or erythorbate reduction of nitrite is potentiated by EDTA salts which have a catalytic effect in the presence of heme pigments and accelerate the curing reaction. It has also been observed (11) that garlic and onion oils, when used at 1500 micrograms per gram of meat slurry, will inhibit toxin production by C. botulinum type A (strain 73A). However, inhibition was incomplete and toxin production by type B and E strains was not inhibited.

Tompson et al. (53) indicated that the ability of nitrite to inhibit C. botulinum growth is affected by iron. Iron added from 0-40 ppm to perishable canned ham cured with 156 ppm of nitrite markedly reduced the antibotulinal efficacy of the nitrite. When 50-500 ppm of the sequestering agent EDTA was added, the effects of iron were nullified.

Goodfellow (17) suggested that acid compounds such as sodium acid pyrophosphate will lower pH and also residual nitrite. Sebranek (43) indicated that this method is also known to provide additional protection against C. botulinum. Sink and Hsu (46) demonstrated that phenolic compounds of a liquid smoke dip process for frankfurters lowered the pH and residual nitrite. Knowles (27) suggested that smoke effects were a combination of pH decrease and direct nitrosation of phenolic compounds to lower residual nitrite.

NITRITE SPARING AGENTS

Various basic approaches have been taken in finding additives that will at least act as sparing agents for nitrite and effectively inhibit growth of C. botulinum. One approach has been to test the few antimicrobial additives that are considered as GRAS by the U.S. Food and Drug Administration. To date, this has met with limited success. Potassium sorbate was found to be equally if not more effective than nitrite in inhibiting C. botulinum growth and toxin production in bacon that had been temperature-abused (25,45). These studies were conducted in our laboratories at Virginia Polytechnic Institute and State University as well as by the USDA and other universities and companies. Even though sorbate is considered as a GRAS substance by the U.S. Food and Drug Administration, there has been some question about the possible allergenic response of consumers to this substance when used in meat products (5). However, this claim has not been fully substantiated.

A “chemical-like” flavor and aroma have been noted among test samples of bacon made with 40 ppm of nitrite and 2600 ppm of potassium sorbate. However, Ivey et al. (25) did not observe such adverse reactions. Additional information (5) has suggested that the aroma may be a low-temperature volatile substance which is more intense very early in the cooking process and that reported reactions may be attributed to the number of bacon samples that the sensory panel members tasted. Control samples were found to produce the same reactions when the same quantities were evaluated.

Another method of inhibiting C. botulinum has centered around processing procedures and alteration of the content of the currently-used curing ingredients. One technique has involved use of a starter culture to lower the pH of bacon during processing, thereby reducing the amount of residual nitrite as well as preventing growth of C. botulinum by acid that is formed (17). This process has not been widely accepted by industry and presents processors with additional processing problems.

The lack of a wide variety of compounds that will inhibit C. botulinum growth and toxin production in meats has led to another approach in finding sparing agents for nitrite. These studies have mainly involved the screening of a wide variety of compounds for their anti-clostridial activity. The p-hydroxybenzoic acid n-alkyl esters have been found to be active in the inhibition of C. botulinum growth (12,42). Other chemicals tested have included the 5-nitrothiazoles (13) and alphatic amines and long chain alphatic amidimides (21).

Herring (19) reported a study that tested the effectiveness of ascorbates in blocking nitrosamine formation. Results indicated that when 1,000-2,000 ppm of ascorbate were added, nitrosamine formation appeared to be blocked. When 250 or 500 ppm of ascorbate were added, nitrosopyrrolidine formation was apparently curbed. However, excessive levels of ascorbate decrease the efficacy of nitrite in bacon due to a more rapid depletion of residual nitrite.

In an attempt to determine the effect of sodium ascorbate and sodium nitrite on toxin formation by C. botulinum in frankfurters, Bowen et al. (7) found that moderate levels of sodium ascorbate, commonly used to enhance the curing process, did not alter the effectiveness of sodium nitrite in inhibition of botulinum toxin formation. These authors did not explain the greater number and earlier development of toxic samples in this study over an earlier experiment conducted by Hustad (22) in the same laboratory.

As another part of the study of the effects of nitrite and ascorbate on botulinal toxin formation in frankfurters and bacon, Bowen and Deibel (8) reported that ascorbate had a significant effect on the nitrite reaction in bacon. These authors also indicated that it is the amount of initial nitrite, rather than the residual amount, which is
significant for control of toxin production. According to Tompkin et al. (52), isoascorbate enhances nitrite inhibition due to its sequestering action instead of its antioxidant or reducing properties similar to ascorbate, cysteine and EDTA. These workers postulated that 0.02% isoascorbate enhances nitrite inhibition of C. botulinum by sequestering a metal ion in the cured meat.

Tompkin et al. (51) found that a moderate level of 200 ppm of ascorbate enhanced the efficacy of nitrite against C. botulinum. Their data revealed that 50 ppm of nitrite plus 200 ppm of isoascorbate inhibited C. botulinum as effectively as did 156 ppm of nitrite alone. These workers suggested that omission of isoascorbate from various product formulations could explain the relative ineffectiveness of nitrite in studies involving perishable cured meats.

Potassium sorbate and sorbic acid were initially used to selectively culture clostridia instead of to inhibit these microorganisms. However, the potential inhibitory effect of sorbate in a meat product has been demonstrated (48,50). A more recent investigation by Pierson et al. (37) indicated that 0.26% potassium sorbate inhibits Staphylococcus aureus when applied in combination with as little as 40 ppm of nitrite (as well as 120 ppm or higher concentration of nitrite). Pierson (36) discovered that addition of sorbate to bacon delays development of toxin produced by C. botulinum. He found that a combination of 0.26% potassium sorbate and 40 ppm of nitrite was equal to or better than 120 ppm of nitrite alone.

Four studies conducted by Sofos et al., and discussed in their thorough review of botulism in cured meats (47), has indicated that 0.2% sorbic acid inhibits botulinum spore germination in mechanically deboned poultry, beef and pork frankfurter emulsions stored at 27 C. It was discovered that 20-156 ppm of nitrite did not affect spore germination but delayed toxin production when 156 ppm was tested. Research by Perry et al. (35) suggested that sorbic acid did not cause off-flavors in poultry when used at recommended levels. However, more work is needed to determine if similar results may be obtained from other meats.

Ivey et al. (25) attempted to reduce the initial nitrite content in cured bacon and maintain safety against botulism. These workers evaluated 0 and 40 ppm of nitrite and 0.13 and 0.26% potassium sorbate. Control samples of commercial bacon formulated with 80 and 120 ppm of nitrite were compared with the test samples. The results indicated that potassium sorbate significantly reduced toxin production. It was discovered that the presence or absence of 40 ppm of nitrite had no significant effect on inhibition by sorbate. Nitrosopyrrolidine formation was reduced with decreased nitrite levels and microbial growth on uninoculated samples was retarded by sorbate (25,41). Ivey and Robach (24) studied the effectiveness of sorbic acid and low nitrite concentrations for inhibition of botulinal growth in a canned comminuted pork product. Toxin production was delayed when 0.2% sorbic acid was added, while 0.1% sorbic acid was ineffective. Inclusion of sodium acid pyrophosphate or sodium hexametaphosphate had a synergistic effect with sorbic acid. Similar effects were observed for comminuted chicken product (40).

Pierson (36) further demonstrated the effectiveness of potassium sorbate as a nitrite-sparing agent. His research revealed that 0.13% potassium sorbate (equivalent to 0.10% sorbic acid) had limited effectiveness, while 0.13% potassium sorbate in combination with 40 ppm of nitrite was more effective. The efficacy of potassium sorbate was increased with a 0.26% concentration and a combination of 0.26% sorbate and 40 or 80 ppm of nitrite was equivalent to or better than 120 ppm of nitrite without sorbate (36) and the resultant product was equal in color and sensory qualities (33).

One group of compounds that has not been extensively screened is the antioxidants. This general group of compounds is widely used in food products. Although Fiddler et al. (15) reported that alpha-tocopherol was a more effective nitrosamine-blocking agent than were the long-chain acetals of ascorbic acid, other lipid-soluble potential agents exist. Some of the more common antioxidants are commonly known as butylated hydroxyanisole (BHA) butylated hydroxytoluene (BHT), tertiary butylhydroquinone (TBHQ), and propylgallate. There have been several reports indicating that the antioxidants have antimicrobial activity. Successful use of ascorbyl palmitate and propylgallate has been reported by Sen et al. (44). Preliminary results (2) indicated that TBHQ may function as a blocking agent of nitrosopyrrolidine formation in bacon.

Chang and Branen (9) found that the 150 ppm of BHA in nutrient broth inactivated Staphylococcus aureus and 400 ppm was lethal to Salmonella typhimurium. Stern et al. (49) reported that the effectiveness of BHA-inhibition of S. aureus was enhanced by addition of sodium chloride and a lowering of the pH of the test medium. An additive effect of BHA and propyl paraben in the inhibition of the growth of S. typhimurium and S. aureus has also been observed. Pierson et al. (48) discovered that BHA at 50 ppm in prerduced Thiotone-Yeast extract was inhibitory to growth of C. botulinum A and B. BHT and propylgallate have been less effective in inhibiting outgrowth of C. botulinum spores than was BHA (42). Sebranek (43) suggested that the combination of alpha-tocopherol and ascorbate is effective by providing reductants for both water-soluble and fat-soluble tissue fractions. Alpha-tocopherol and ascorbyl palmitate have been found to most effectively block formation of N-nitrosopyrrolidine during frying of bacon.
While ascorbyl palmitate has a similar action in reducing nitrosopyrrolidine production, no synergistic effect between it and alpha-tocopherol has been noted when the two antioxidants are combined. Since it has been shown that formation of nitrosopyrrolidine occurs mainly in the fat of bacon (14,34), lipid-soluble antioxidants such as alpha-tocopherol should be more effective than water-soluble ascorbate and erythorbate.

Mergens and Newmark (30) have studied and reported on the use of alpha-tocopherol in model systems to prevent nitrosamine formation. Alpha-tocopherol was evaluated as a nitrite-scavenger and was found to react with nitrosating agents in both lipophilic and aqueous environments. These workers reported that samples containing 500 ppm of ascorbate and 500 ppm of alpha-tocopherol showed nitrosopyrrolidine concentrations ranging from 3 to 6 ppb in the edible bacon portion.

A study conducted by the American Meat Institute (1) clearly demonstrated that alpha-tocopherol was effective in blocking formation of n-nitrosopyrrolidine in bacon during drying. In this work, n-nitrosopyrrolidine concentrations were lowered in samples containing reduced nitrite (120 ppm) and alpha-tocopherol to an average of less than 2 ppb. Intake of alpha-tocopherol from bacon containing 500 ppm has been estimated to be 5% of the U.S. Recommended Daily Dietary Allowance. These workers have indicated that 70-80% of alpha-tocopherol is retained during refrigerated storage and about 70% after frying of bacon slices.

Parahydroxybenzoic acid esters (parabens) which are used as antimicrobial agents in the food industry have been reported to be effective inhibitors of C. botulinum when used with low levels of nitrite in microbiological media (42). However, parabens have limited effectiveness for this application. Radiation sterilization to destroy toxin-producing C. botulinum spores offers another alternative to allow reduction or elimination of nitrite but does not appear to have the commercial potential of other methods.

Therefore, it seems that there are several possible compounds, including antioxidants, that might have a potential use as antclostridial additives in cured meats. The effect of the antioxidants is thought to be related to the phenolic base of many of these compounds. There needs to be a more extensive screening of these potential inhibitors in both bacteriological media and meat systems containing other cure ingredients.

A method proposed for USDA approval as a safe alternative to the current procedure of 120 ppm of ingoing sodium nitrite with 550 ppm of ascorbate in bacon is use of a lactobacillus culture (17). The culture reduces meat pH, which ultimately reduces residual nitrite in the cured product and also renders the nitrite more effective. With less residual nitrite, nitrosamine formation is reduced.

SUMMARY

Nitrite-sparing agents that have been discussed should be considered for cured meats in the future, since most of the agents that were discussed are "naturally occurring" compounds with preservation qualities. These compounds will contribute to the industry by permitting lower nitrite levels with less potential for nitrosamine formation. However, this approach should provide protection against botulism and maintain the desired cured meat color and flavor without a major change in current processing techniques.

REFERENCES


Fung and Cox, con't. from p. 880


