

Nitrite Reaction Rates with Substrates for Meat Curing in a Model System

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Summary and Implications

Because one of the alternative processes for production of natural or organic cured meat products utilizes a slow release of nitrite from nitrate to achieve the cure, the reaction rates of nitrite with myoglobin and cysteine in a model system were assessed to determine if the rate of addition of nitrite has potential to alter the end products produced by curing. This is potentially important for natural and organic cured meats because the end products of nitrite reactions determine cured meat properties including color and antimicrobial protection. Results of this study showed that myoglobin is preferentially nitrosylated by nitrite before cysteine. Once maximum nitrosylation of myoglobin was achieved, then nitrite reacted with cysteine as well. These results suggest that the amount of nitrite produced may affect end products of meat curing based on the substrates used in this study, but the rate of addition of nitrite is not important. Thus, for natural and organic curing achieved by the alternate process involving nitrate, it may be more important to increase the amount of nitrite produced from nitrate rather than increasing the rate of nitrite production. However, additional studies with other substrates involved with nitrite in meat curing should be conducted.

Introduction

Meat curing involves a highly complex series of reactions involving nitrite and a wide range of substrates that produce the end products responsible for cured meat properties. Nitrite is typically added as an ingredient during formulation of cured meats. However, recent development of natural and organic processed meats has required use of alternative curing processes because USDA regulations do not permit direct addition of nitrite to these products. One of the alternative processes that have been developed utilizes celery powder as a natural source of nitrate and a bacterial starter culture that provides the reduction of nitrate to nitrite to achieve curing reactions. Because production of nitrite by bacterial action is slow, and nitrite reactions with meat components range from slow to fast, we hypothesized that slow production of nitrite in natural and organic products would favor a greater proportions of end products from substrates that react quickly with nitrite at the expense of those that are relatively slow to react. Changing the relative amounts of end products of curing could have

implications for cured meat properties such as microbial control, a property that has been reported to be less effective in natural and organic products than in conventionally-cured meats. To test this hypothesis, myoglobin and cysteine were used as substrates in a model meat curing system to assess the effect of the rate of nitrite addition on reactions of nitrite with these two substrates.

Materials and Methods

The model system used for this study was composed of 0.029 mM equine myoglobin (chosen because it does not contain cysteine) and 5.06 mM cysteine in 0.1 M phosphate buffer solutions at either pH 5.6 or pH 7.4. These myoglobin and cysteine concentrations are approximately half those found in fresh ham and allowed for spectrophotometric measurement of color changes in the solutions as a result of nitrite addition. Nitrite solutions were prepared to give the equivalent of 0, 10, 25, 50, 100, 150, 200 and 500 parts per million (ppm) ingoing nitrite concentration when added to the substrate solutions. To simulate the traditional cure, all of the nitrite solution (5 ml) was added to the substrate solution at the beginning of the experiment. To simulate the natural cure with slow release of nitrite by bacteria, 1 ml of the nitrite solution was added to the substrates at the beginning of the experiment, samples were held at 35°C, and an additional 1 ml of nitrite solution was added every 10 minutes until the total reached 5 ml. After 60 minutes, all samples were heated to 75°C to simulate final product cooking of cured meats. Loss of sulfhydryl groups of cysteine due to reaction with nitrite were measured by Ellman's reagent and myoglobin reaction with nitrite was determined by measuring the concentration of the nitrosohemochromagen pigment color produced as a result of the reaction of nitrite with myoglobin. The reaction rates of nitrite with myoglobin and cysteine alone, and in combination, were evaluated.

Results and Discussion

The reaction of nitrite with myoglobin was fast and reached a maximum at about 25 ppm of nitrite after which the reacted pigment concentration was similar as nitrite concentration increased (Figure 1). There was no difference between the different rates of nitrite addition. Nitrite reaction with cysteine did not occur until the myoglobin reaction plateaued at about 25 ppm (Figure 2) but then increased (as measured by decreased sulfhydryl groups) as nitrite concentrations increased. Again, as for myoglobin, there was no effect of the rate of nitrite addition on reaction of nitrite with cysteine. These results suggest that myoglobin is nitrosylated more quickly than cysteine is nitrosated which would explain the formation of cured color

prior to the reaction of nitrite with cysteine. This means that slow release of nitrite as occurs in natural curing does not affect or shift the amount of nitrite between reaction intermediates or reaction end products, and it may be more important to increase the amount of ingoing nitrite in natural and organic cured meats rather than increasing the rate of nitrite formation in these products to achieve desired cured

meat properties. However, this may not be true for other nitrite reaction substrates found in meat. Only two potential substrates were utilized in this study. Future studies should include other substrates – including other added ingredients – to investigate this phenomenon.

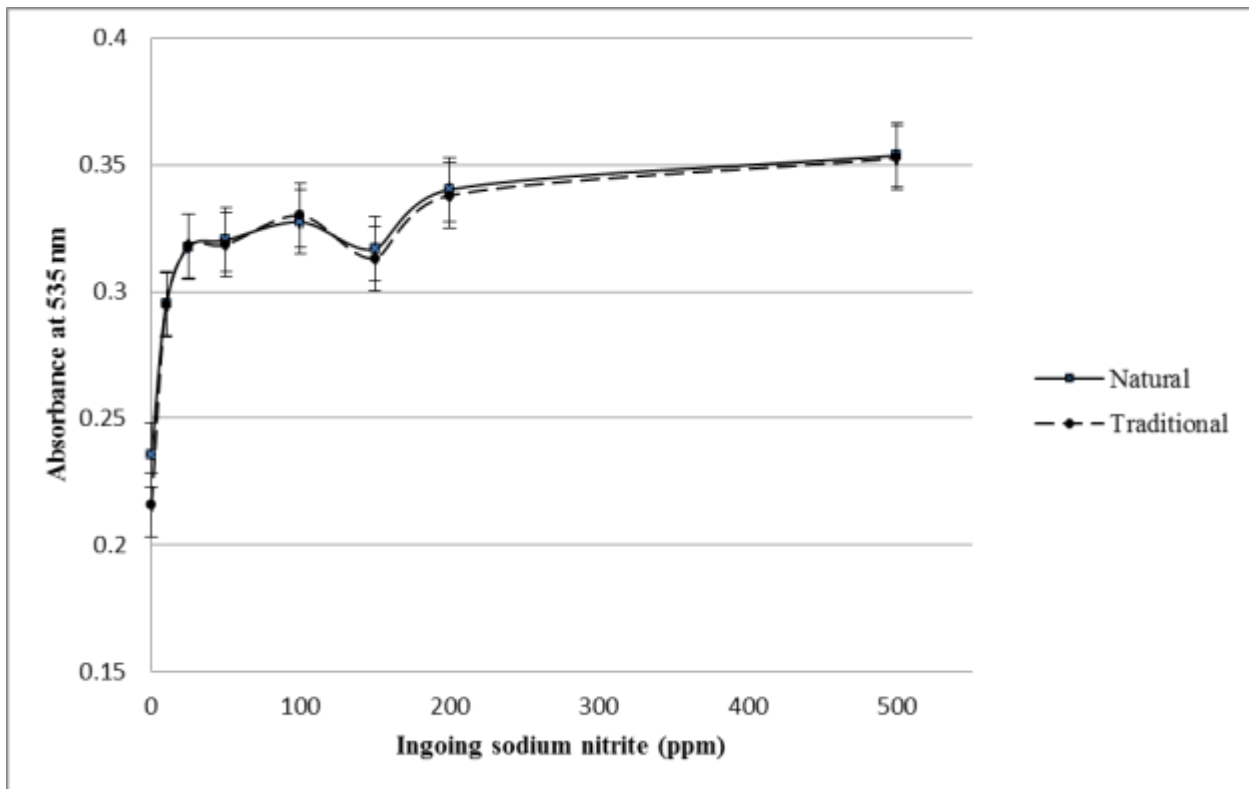


Figure 1. Absorbance at 535 nm of cysteine-plus-myoglobin model system as an indicator of nitrosylhemochromogen formation. Natural = sodium nitrite solution added in 1 ml increments for simulated bacterial reduction of nitrate to simulate natural curing. Traditional = entire sodium nitrite solution added at beginning to simulate traditional curing.

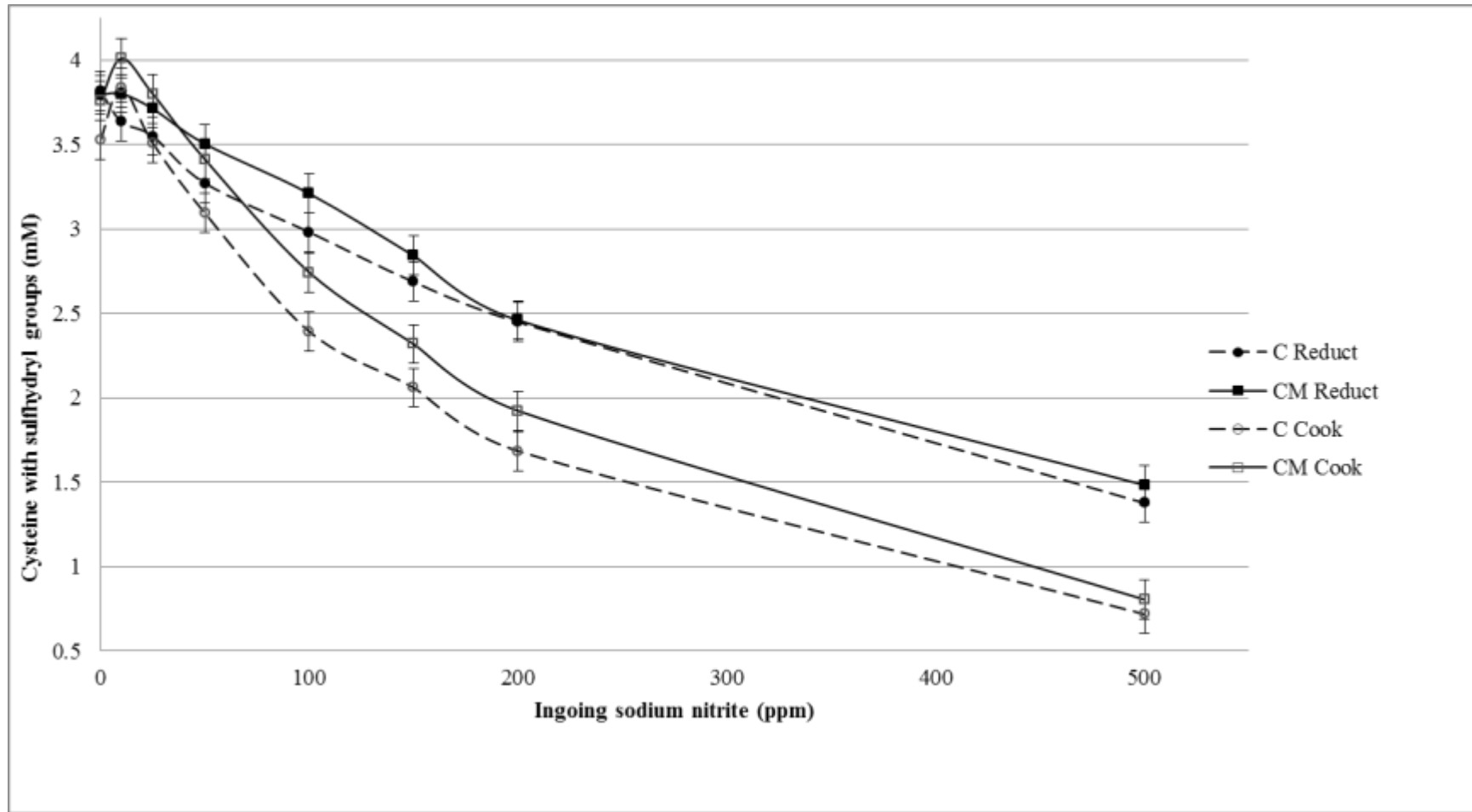


Figure 2. Concentration of cysteine with intact sulfhydryl groups. C Reduct = Cysteine-only model evaluated following simulated bacterial reduction; CM Reduct = Cysteine-plus-myoglobin model evaluated following simulated bacterial reduction; C Cook = Cysteine-only model evaluated following cooking simulation; CM Cook = Cysteine-plus-myoglobin model evaluated following cooking simulation.