This is to certify that the

thesis entitled

The Use of Nitrite-Sorbate Combinations
In Cured Poultry Products

presented by

Diane Marie Bussey

has been accepted towards fulfillment
of the requirements for

M.S. degree in Food Science

J. J. Gray
Major professor

Date 9/15/81

O-7639

# THE USE OF NITRITE-SORBATE COMBINATIONS IN CURED POULTRY PRODUCTS

By

Diane Marie Bussey

A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Food Science
and Human Nutrition

1981

ABSTRACT

THE USE OF NITRITE-SORBATE COMBINATIONS
IN CURED POULTRY PRODUCTS

By

Diane Marie Bussey

Sensory and chemical analyses of turkey bologna and turkey ham prepared with various levels of sodium nitrite (0 to 156 mg/kg) indicated that the introduction of 40 mg/kg of nitrite into either product provided an organoleptically acceptable sample that contained no detectable N-nitrosamines, exhibited reduced TBA values and had Hunter color results which were not significantly ($p < 0.01$) different from the reference (156 mg/kg of nitrite). The same type of poultry products were subsequently manufactured with various combinations of potassium sorbate and reduced levels of nitrite (40 and 60 mg/kg). Samples prepared without nitrite or with sorbate alone (0.26 and 0.39%) were unacceptable, but the combination of 40 mg/kg of nitrite and 0.26% sorbate provided products that contained no detectable N-nitrosamines and which exhibited sensory, TBA and Hunter color values comparable to the reference. Furthermore, 60 mg/kg of nitrite combined with 0.26% sorbate was found to be as effective as 156 mg/kg of nitrite in inhibiting Clostridium botulinum.

As a token of my appreciation,
this thesis is dedicated to my husband:

Michael P. Bussey

The combination of his encouragement, support, advice,
patience and limitless love was a major asset,
in the completion of my research
and thesis work.

## ACKNOWLEDGEMENTS

I wish to thank my major professor, Dr. J. I. Gray, for his time, guidance and encouragement throughout the course of this research. I would also like to thank my committee members, Dr. L. E. Dawson, Dr. A. M. Pearson, Dr. J. F. Price and Dr. M. J. Zabik for their review of this thesis. A special note of appreciation is extended to Dr. J. F. Price for the use of the meats laboratory and for his time, guidance and patience during the formulation and preparation of the poultry products. The statistical advice from Dr. J. L. Gill and the technical assistance by Ms. S. L. Cuppett, Mr. D. Morris and Mrs. K. L. Uebersax were most appreciated. I would like to thank the various members of my lab group for their contributions during the preparation and sensory analysis of my products. I also appreciate the botulinal toxin assays completed by Mrs. M. Dynnik and the N-nitrosamine analyses of my products by Mr. M. C. Robach and Mr. J. L. Owens from the Monsanto Industrial Chemical Company, St. Louis, MO. The financial assistance from the National Institute of Health and the Ralston Purina Company were extremely helpful. Lastly, a heartfelt thank-you to my husband, Michael, who continually reminded me about the "light at the end of the tunnel".

TABLE OF CONTENTS


| | Page |
|---|---|
| LIST OF TABLES | vi |
| LIST OF FIGURES | ix |
| I. INTRODUCTION | 1 |
| II. LITERATURE REVIEW | 5 |
| A. FUNCTIONS OF NITRITE | 5 |
| 1. Color Fixation | 5 |
| 2. Flavor | 14 |
| a. Cooked, uncured meat flavor | 14 |
| b. Cured meat flavor | 17 |
| c. Warmed-over flavor | 21 |
| 3. Antibotulinal Activity | 24 |
| B. NITRITE AND N-NITROSAMINE FORMATION | 30 |
| C. NITRITE SUBSTITUTES | 37 |
| D. SORBATE | 45 |
| III. TURKEY BOLOGNA STUDY | 56 |
| A. EXPERIMENTAL | 56 |
| 1. Bologna Preparation | 56 |
| 2. Organoleptic Analysis | 58 |
| 3. Analytical Methods | 59 |
| 4. Statistical Analysis | 60 |
| 5. N-Nitrosamine Analysis | 61 |
| 6. Clostridium botulinum Study | 63 |
| B. RESULTS AND DISCUSSION | 65 |
| 1. Test 1: Determination of Minimum Nitrite Level | 65 |
| a. Organoleptic analysis | 65 |

Page

b. Proximate analysis and pH ..... 84
c. Residual nitrite ..... 85
d. Hunter colorimetry ..... 87
e. TBA values ..... 89
f. N-Nitrosamine analysis ..... 91
2. Test 2: Nitrite-Sorbate Combinations ..... 93
a. Organoleptic analysis ..... 93
b. Proximate analysis and pH ..... 109
c. Residual nitrite ..... 111
d. Hunter colorimetry ..... 113
e. TBA values ..... 114
f. N-Nitrosamine analysis ..... 116
g. Clostridium botulinum study ..... 117

IV. TURKEY HAM STUDY ..... 120

A. EXPERIMENTAL ..... 120

1. Ham Preparation ..... 120
2. Organoleptic Analysis ..... 122
3. Analytical Methods ..... 123
4. Statistical Analysis ..... 123
5. N-Nitrosamine Study ..... 123
6. Clostridium botulinum Test ..... 123

B. RESULTS AND DISCUSSION ..... 124

1. Test 1: Determination of Minimum Nitrite Level .... 124
a. Organoleptic analysis ..... 124
b. Proximate analysis and pH ..... 141
c. Residual nitrite ..... 142
d. Hunter colorimetry ..... 144
e. TBA values ..... 145
f. N-Nitrosamine analysis ..... 147
2. Test 2: Nitrite-Sorbate Combinations ..... 149
a. Organoleptic analysis ..... 149
b. Proximate analysis and pH ..... 166
c. Residual nitrite ..... 168
d. Hunter colorimetry ..... 169
e. TBA values ..... 170
f. N-Nitrosamine analysis ..... 172
g. Clostridium botulinum study ..... 172

V. SUMMARY AND CONCLUSIONS ..... 175

BIBLIOGRAPHY ..... 178

APPENDIX - SENSORY PANEL EVALUATION FORM ..... 197
TABLES A-1 THROUGH A-64 ..... 202

LIST OF TABLES


| TABLE | | Page |
|---|---|---|
| 1. | Botulism outbreaks | 25 |
| 2. | Target nitrite levels added to turkey bologna for Test 1. | 57 |
| 3. | Target nitrite-sorbate concentrations added to turkey bologna for Test 2 | 57 |
| 4. | Cooking schedule for turkey bologna (Tests 1 and 2) | 58 |
| 5. | Linear correlation and regression of various sensory attributes of turkey bologna containing various nitrite treatments | 84 |
| 6. | Proximate analysis of raw turkey thigh meat and non-nitrite treated turkey bologna | 85 |
| 7. | pH values for turkey bologna prepared with various levels of sodium nitrite and stored at 4°C over three weeks | 86 |
| 8. | Residual nitrite analysis of turkey bologna prepared with various nitrite levels and stored at 4°C | 86 |
| 9. | Relationship between Hunter color values and subjective evaluation of turkey bologna color | 89 |
| 10. | Relationship between TBA values and subjective flavor and odor evaluations of turkey bologna | 91 |
| 11. | Presumptive N-nitrosamine levels (μg/kg in turkey bologna prepared with various levels of sodium nitrite | 92 |
| 12. | Linear correlation and linear regression of various sensory attributes for turkey bologna containing nitrite-sorbate treatments | 110 |
| 13. | Proximate analysis of raw turkey thigh meat and non-nitrite treated turkey bologna | 110 |
| 14. | pH values for turkey bologna prepared with various nitrite-sorbate treatments and stored at 4°C | 111 |

TABLE Page

15. Residual nitrite analysis of turkey bologna prepared with various nitrite-sorbate combinations and stored at 4°C.... 112

16. Relationship between Hunter color values and the subjective evaluation of turkey bologna color..... 114

17. Relationship between TBA values and subjective flavor and odor evaluations for turkey bologna..... 115

18. Presumptive N-nitrosamine levels (μg/kg) in turkey bologna prepared with various nitrite-sorbate treatments..... 116

19. Influence of nitrite-sorbate combinations on the Clostridium botulinum toxinogenesis in turkey bologna held at 27°C over 28 days..... 118

20. Target nitrite levels for turkey ham (Test 1)..... 121

21. Target nitrite-sorbate levels for turkey ham (Test 2)..... 121

22. Cooking schedule for turkey hams (Tests 1 and 2)..... 122

23. Linear correlation and linear regression of various sensory attributes of turkey hams containing various levels of sodium nitrite..... 140

24. Proximate analysis of turkey thigh meat and nonnitrite treated turkey ham..... 141

25. pH values for turkey ham prepared with various levels of nitrite and stored at 4°C over three weeks..... 142

26. Residual nitrite analysis of turkey ham prepared with various levels of nitrite and stored at 4°C over a three week period..... 143

27. Relationship between Hunter values and the subjective evaluations for turkey ham color..... 145

28. Relationship between TBA values and the subjective evaluations for turkey ham flavor and odor..... 147

29. Presumptive N-nitrosamine levels (μg/kg) in turkey ham prepared with various levels of sodium nitrite..... 148

30. Linear correlation and linear regression between various sensory attributes for turkey hams prepared with nitrite-sorbate combinations..... 166

TABLE Page

31. Proximate analysis of turkey thigh meat and nonnitrite treated turkey ham..... 167

32. pH values for turkey ham prepared with various nitrite-sorbate treatments and stored at 4°C for three weeks..... 167

33. Residual nitrite analysis of turkey ham cured with various nitrite-sorbate combinations and stored at 4°C over a three week period..... 168

34. Relationship between Hunter values and the subjective evaluation of turkey ham color..... 170

35. Relationship between TBA values and subjective flavor and odor evaluations of turkey hams..... 171

## LIST OF FIGURES


| FIGURE | | Page |
|---|---|---|
| 1. | Some of the possible curing reactions which occur with the use of nitrite (Kemp, 1974)..... | 7 |
| 2. | Effect of various nitrite levels on the mean panel scores for turkey bologna color during storage at 4°C... | 67 |
| 3. | Effect of various nitrite levels on the mean panel scores for turkey bologna flavor during storage at 4°C.. | 70 |
| 4. | Effect of various nitrite levels on the mean panel scores for turkey bologna odor during storage at 4°C.... | 72 |
| 5. | Effect of various nitrite levels on the mean panel scores for turkey bologna off-flavor during storage at 4°C..... | 76 |
| 6. | Effect of various nitrite levels on the mean panel scores for turkey bologna off-odor during storage at 4°C..... | 78 |
| 7. | Effect of various nirrite levels on the mean panel scores for turkey bologna overall acceptability during storage at 4°C..... | 81 |
| 8. | Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna color during storage at 4°C..... | 95 |
| 9. | Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna flavor during storage at 4°C..... | 98 |
| 10. | Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna odor during storage at 4°C..... | 100 |
| 11. | Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna off-flavor during storage at 4°C..... | 103 |

FIGURE Page

12. Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna off-odor during storage at 4°C..... 105

13. Effect of various nitrite-sorbate treatments on the mean panel scores for the overall acceptability of turkey bologna during storage at 4°C..... 108

14. Effect of various nitrite levels on the mean sensory scores for turkey ham color during storage at 4°C..... 126

15. Effect of various nitrite levels on the mean sensory scores for turkey ham flavor during storage at 4°C..... 128

16. Effect of various nitrite levels on the mean sensory scores for turkey ham odor during storage at 4°C..... 130

17. Effect of various nitrite levels on the mean sensory scores for turkey ham off-flavor during storage at 4°C.. 134

18. Effect of various nitrite levels on the mean panel scores for turkey ham off-odor during the storage at 4°C..... 136

19. Effect of various nitrite levels on the mean sensory scores for the overall acceptability of turkey hams during storage at 4°C..... 139

20. Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham color during storage at 4°C..... 151

21. Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham flavor during storage at 4°C..... 154

22. Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham odor during storage at 4°C..... 156

23. Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham off-flavor during storage at 4°C..... 159

24. Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham off-odor during storage at 4°C..... 161

FIGURE Page

25. Effect of various nitrite-sorbate treatments on the mean sensory scores for the overall acceptability of turkey ham stored at 4°C..... 164

# I. INTRODUCTION

The curing of meat is as old as the desire of primitive man to preserve a portion of one day's kill for later use (Lechowich et al., 1978). The use of salt in meat preservation dates to as early as 3000 B.C. in Mesopotamia. Although such desert salts contained nitrate and borax as impurities, it was not until Roman times that the red color and desirable flavor of the meat product was attributed to nitrate (Binkerd and Kolari, 1975; Sofos et al., 1979a). By the early 20th century, meat curing was transformed from an art to a science when chemists were employed by the meat industry. Development of curing methods, such as dry cure, wet and pickle cure combinations and pumping occurred during this time. Along with the traditional salt and nitrate, other ingredients such as spices, phosphates, ascorbate and other reductants were incorporated into curing formulations. Another achievement of this period was the recognition that the nitrate added to meat was converted to nitrite through bacterial reduction, and that nitrite instead of nitrate was responsible for cured meat color development (Binkerd and Kolari, 1975; Lechowich et al., 1978; Sofos et al., 1979a). In 1925, research revealed that the direct addition of nitrite (instead of nitrate) produced a more uniform color in the product and decreased chances of spoilage (Kerr et al., 1926). The USDA then authorized the use of ¼ ounce of sodium nitrite per 100 pounds of meat (156 mg/kg), with the finished product containing no more than 200 mg/kg of sodium nitrite (Binkerd and Kolari, 1975).

Over the years, experience and scientific knowledge have indicated that nitrite has several pronounced effects on meat (Sofos et al., 1979a). It (a) produces the characteristic cured meat color and flavor, (b) has antioxidant activities which prevent "warmed-over" flavor, and (c) retards Clostridium botulinum growth and toxin production which can occur if the product is mishandled and temperature-abused. Despite the significant contribution of nitrite to the aesthetic qualities and microbiological safety of meat products, its continued use in meat curing has become a controversial issue due to reports showing nitrites added to certain meats will react with various amino compounds to form carcinogenic N-nitrosamines (Sofos et al., 1979a). These compounds have been found sporadically in cured "red" meat such as hams, bologna, frankfurters and similar products, usually in amounts below 25 μg/kg (Gray and Randall, 1979). Bacon, on the other hand, presents a more serious problem since N-nitrosopyrrolidine (NPYR) has been isolated consistently from cooked bacon (Gray, 1976). As a consequence, the Expert Panel on Nitrites, Nitrates and Nitrosamines in its final report to the Secretary of Agriculture recommended lowering the permitted levels of nitrite or eliminating it completely from some cured meat products (Anon., 1978a).

A risk-benefit controversy then developed for industrial, research and governmental factions. The excellent botulism safety record of commercially processed cured meat is primarily attributed to inclusion of nitrite in the formulation (Sofos et al., 1979a), but the mutagenic, teratogenic and carcinogenic properties of many N-nitroso compounds found in cured meats have also been frequently documented. How can the absolute danger of botulism be balanced against the potential threat of cancer? In response, the antibotulinal activity of many nitrite

supplements or replacers has been assessed. Recently, some reports dealing with the use of sorbic acid or its potassium salt in preventing botulinal toxicity in cured meats have been published (Ivey et al., 1978; Robach et al., 1978a; Sofos et al., 1979c). However, studies of this type are few and limited.

A group of processed meat products which have witnessed increased consumption and popularity in the past five years is the variety of cured poultry meats, such as turkey ham, turkey bologna and chicken frankfurters. In contrast to raw, fresh poultry which will spoil or develop unpleasant odors within 10-14 days of slaughter, cured poultry has a much longer shelflife (Bauermann, 1979). The contribution of nitrite to the color, flavor and botulinal safety of such products has not been evaluated. Nor has it been determined whether the addition of nitrite to these products may result in the formation of N-nitroso compounds during processing and storage. It is also inappropriate to apply cured red meat information to poultry meat substrates. The questionable use of nitrite in poultry is compounded since at the present time, the poultry industry is not protected under the "grandfather" clause of the Delaney Cancer Amendment, and as such, the poultry industry cannot use that as the means of justifying the continued use of nitrite in poultry products. There are no Food and Drug Administration regulations that independently authorize the commercial use of nitrites and nitrates in poultry products, either as food additives or as color additives; nor has the Food and Drug Administration itself issued a prior sanction permitting their use (D.H.E.W., 1977).

Therefore, the controversy over the use of nitrite in cured poultry products, the general lack of information regarding the presence of N-nitrosamines in these products, the indications of a possible sorbic acid effect in inhibiting botulinal toxin production, and the increased consumption of cured poultry products in the United States justify an investigation into the use of nitrite-sorbate combinations in the manufacture of cured poultry products (specifically, turkey bologna and turkey ham).

The primary objectives of this research project were as follows: (1) to determine the minimal nitrite level that results in an acceptable cured poultry product according to sensory, chemical and N-nitrosamine analyses; (2) to determine the effects of potassium sorbate and reduced nitrite levels (alone or in combination) on the subjective (sensory) evaluation and on the objective (chemical) analyses of turkey bologna and turkey ham; (3) to determine the effects of potassium sorbate and reduced nitrite levels (alone or in combination) in cured poultry products on the Clostridium botulinum toxigenesis during temperature abuse at 27°C, and (4) to determine the effects of potassium sorbate and reduced nitrite levels on the formation of volatile N-nitrosamines in turkey bologna and turkey ham.

## II. LITERATURE REVIEW

### A. FUNCTIONS OF NITRITE

#### 1. Color Fixation

The bright, cherry-red appearance from the oxymyoglobin in fresh red muscle and the reddish-pink hue of denatured nitrosylmyohemochrome in cured meat products are recognized as being the overriding point-of-purchase quality attributes. Good appearance does not necessarily predict good texture and flavor (or vice versa): nevertheless, the shopper tends to associate this red color in the supermarket display cases with the expectation of wholesome eating enjoyment (Jeremiah et al., 1972; Giddings, 1977a).

As reviewed by Fox (1966), Clydesdale and Francis (1971), Govindarajan (1973), and Giddings (1974, 1977a,b), the color of both fresh and cured meat products is primarily attributed to the hemeprotein pigment, myoglobin. Figure 1 portrays the dynamic equilibrium between the various forms of myoglobin and demonstrates that the resulting hue of a meat product is dependent upon the oxidation state of the heme iron in the pigment and the type of functional group on the sixth ligand of the iron (Fox, 1966). The color of raw or fresh muscle tissue, such as beef or pork, is due to the dark red pigment, myoglobin (Mb); the cherry-red pigment, oxymyoglobin ($O_2Mb$) and the brown pigment, metmyoglobin (MMb) (Reith and Szakaly, 1967a; Clydesdale and Francis, 1971). Many factors influence the stability of these pigments (Fox, 1966;

Figure 1. Some of the possible curing reactions which occur with the use of nitrite (Kemp, 1974).

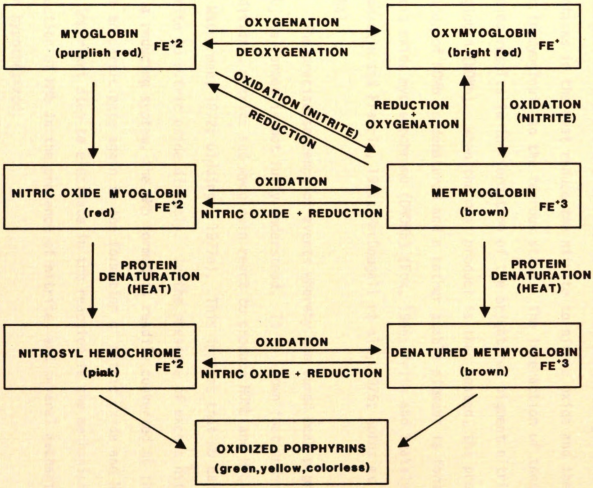

MYOGLOBIN
(purplish red) FE+2
OXYGENATION
DEOXYGENATION
OXYMYOGLOBIN
(bright red) FE+
OXIDATION (NITRITE)
REDUCTION
REDUCTION + OXYGENATION
OXIDATION (NITRITE)
NITRIC OXIDE MYOGLOBIN
(red) FE+2
OXIDATION
NITRIC OXIDE + REDUCTION
METMYOGLOBIN
(brown) FE+3
PROTEIN DENATURATION (HEAT)
PROTEIN DENATURATION (HEAT)
NITROSYL HEMOCHROME
(pink) FE+2
OXIDATION
NITRIC OXIDE + REDUCTION
DENATURED METMYOGLOBIN
(brown) FE+3
OXIDIZED PORPHYRINS
(green,yellow,colorless)

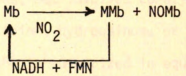
Figure 1

Clydesdale and Francis, 1971; Giddings, 1977a,b) and it is well-known that they are not at all stable when the muscle tissue is heated (Reith and Szakaly, 1967a). To obtain a more stable red pigment in heated commercial meat products, nitrite is added before heating. Biochemical reactions in the meat reduce the nitrite to nitric oxide and the heme iron in myoglobin to the ferrous state. The interaction of these two species results in the formation of the bright red pigment nitric oxide myoglobin (NOMb). When the meat product is then heated, the protein portion of NOMb is denatured and a rather stable pigment is formed, named nitric oxide myohemochrome (DNOMb) (Fox, 1966; Reith ahd Szakaly, 1967a; Clydesdale and Francis, 1971; MacDougall et al., 1975; Sofos et al., 1979a).

The precise sequence of events whereby the cured meat pigment, NOMb, is formed is not fully understood. It is known that under anaerobic conditions, nitrite and myoglobin react to produce NOMb and MMb (Wolff and Wasserman, 1972; Giddings, 1977a). This implies that Mb can reduce nitrite to nitric oxide directly. In the presence of excess nitrite and a reducing system, the MMb formed is readily converted to the reduced form to participate again in the formation of nitric oxide and NOMb. The important step to elucidate in the reaction is the mechanism of the reduction of MMb in the presence of nitrite, and several mechanisms have been hypothesized.

The coenzyme systems, NADH (reduced nicotinamide dinucleotide) or NADPH (reduced nicotinamide adenine dinucleotide phosphate) plus FMN (flavin mononucleotide), was studied by Koizumi and Brown (1971) and was observed to readily produce NOMb from MMb. FAD (flavin adenine

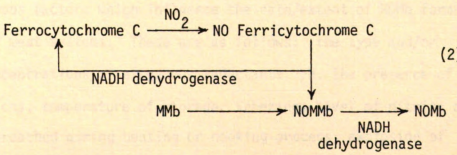
dinucleotide) and riboflavin were also effective for the formation of NOMb by NADH, but in the absence of Mb, the system did not reduce the nitrite to nitric oxide. This mechanism for the formation of NOMb is given in equation (1)




(1)

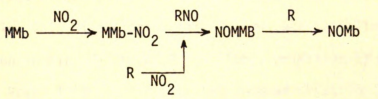
Koizumi and Brown (1971) also studied a model enzyme reducing system, diaphorase-methylene-blue-NADH, which was effective in the formation of NOMb, but in the absence of Mb did not reduce nitrite. This proposed mechanism differs radically from the chemical scheme or enzymatic reactions proposed by others.

The participation of endogenous enzymes of the mitochondria in the reduction mechanism has also been studied (Walters and Taylor, 1963). The system summarized in equation (2) showed that NOMb formation can proceed to nitric oxide metmyoglobin (NOMMb) by transfer of nitric oxide from nitric oxide ferricytochrome C. Nitric oxide ferricytochrome C is formed from the reaction of nitrite on ferrocytochrome C and the cytochrome system is cycled by an NADH dehydrogenase. The NOMMb formed is then reduced by the NADH dehydrogenase.




(2)

Fox and Thomson (1963) showed that after MMb was formed by the action of nitrite, it formed the ionic complex metmyoglobin-nitrite ($MMb\text{-}NO_2$) which then reacted with a semistable nitric oxide reductant intermediate to give NOMMb, which then reduced to NOMb. This scheme does not rely on enzymes, but involves coenzymes and/or other reducing compounds, such as cysteine, hydroquinone or ascorbic acid. This scheme (Fox and Ackerman, 1968) is summarized in equation (3), where R represents a naturally occurring or added reducing compound.




(3)

This mechanism is advantageous because it allows color formation before and after the product is cooked, while heating would eliminate the possibility of the enzyme-coupled reductions suggested by Koizumi and Brown (1971) and by Walters and Taylor (1963). Ascorbic acid has been reported to be a very effective reductant in this capacity, and therefore is usually incorporated into cure formulations to accelerate the development and increase the stability of cured meat color (Watts and Lehmann, 1965; Fox, 1966; Fox et al., 1967).

All of these hypothesized mechanisms must be considered in light of the numerous factors which influence the rate/extent of NOMb formation in model and meat systems. These are as follows: the type and/or relative concentrations of exogenous reductants, pH, the presence of salt/metal ions, temperature of storage, freezing, level of nitrite added, temperature reached during heating or cooking process, exclusion of

oxygen during formulation, and the original pigment concentration in the meat (Weiss et al., 1953; Siedler and Schweigert, 1959; Watts and Lehmann, 1965; Giddey, 1966; Fox et al., 1967; Reith and Szakaly, 1967a,b; Acton et al., 1979; Renerre and Rougie, 1979).

Once formed, the complex of myoglobin and nitric oxide is very stable in the absence of oxygen. In the presence of oxygen, which rapidly oxidizes free nitric oxide to nitrite, the stability of the complex is limited by the rate of nitric oxide dissociation since oxygen does not react directly with the bound nitric oxide (Giddings, 1977a,b). On the whole, this dissociation rate is low, but is believed to occur by autoxidation in air (Walsh and Rose, 1956), oxidation by nitrous acid (Walsh and Rose, 1956), lipid peroxide-induced oxidation(Younathan and Watts, 1959), or by photocatalyzed oxidation (Walsh and Rose, 1956; Bailey et al., 1964). The underlying principle for all such mechanisms of nitric oxide-heme dissociation is believed to involve the withdrawal of electron density from iron to porphyrin, thus weakening the Fe-NO bond. The NO group dissociates leaving the iron susceptible to oxidation by the "electronegative groups" present in the medium (Tarladgis, 1962). Such color loss is believed to be delayed by providing stronger reducing conditions in the medium (Tarladgis, 1962; Bailey et al., 1964; Reith and Szakaly, 1967a; Lin et al., 1980), incorporating nitrite in excess of the Mb level (Walsh and Rose, 1956; Tarladgis, 1962; Reith and Szakaly, 1967a,b), avoiding exposure to any kind of energy causing electronic excitation (Walsh and Rose, 1956; Tarladgis, 1962), replacement of NO-based curing salts with nitrogenous compounds possessing strong electron-donor power (Siedler and Schweigert, 1959; Tarladgis, 1962),

the elimination of oxygen during storage (Fox, 1966; Reith and Szakaly, 1967b), use of packaging films with low oxygen permeability (7 ml $O_2/m^2/24$ h) combined with a maximum (686-737 mm Hg) initial vacuum levels (Kraft and Ayres, 1954; Lin and Sebranek, 1979; Lin et al., 1980), and/or increasing the pH of the product (Walsh and Rose, 1956; Bailey et al., 1964; Reith and Szakaly, 1967a).

It is widely recognized that only a small fraction of the nitrite added to a meat product is utilized for color fixation. In fact, only 3 mg/kg nitrite will provide a 50% conversion of Mb to NOMb and result in adequate color production (MacDougall et al., 1975). However, at least 25 mg/kg nitrite is usually necessary to provide a stable color due to the many factors (previously discussed) which influence the NOMb stability, and also due to the reaction of nitrite with other meat components, such as sulfhydryl or amino groups (Cassens et al., 1974; Woolford and Cassens, 1977; Cassens et al., 1979). Kerr et al. (1926) noted that incomplete color formation was due to insufficient nitrite penetration into the meat and/or due to unusually low myoglobin concentrations. This is exemplified by the fact that the minimum level of nitrite necessary for acceptable color varies with the type of meat product, method of preparation and with the presence of reductants such as ascorbate (MacDougall et al., 1975; Sofos et al., 1979a).

For dry-cured hams, Kemp et al. (1974, 1975) and Eakes and Blumer (1975a) reported that the presence of nitrite, nitrate or their combination resulted in an improved, more desirable color relative to the brownish-gray hue observed in the salt and sugar-treated (control) sample. In both dry-cured hams and pork loins, 70 mg/kg nitrite and/or

nitrate provided significantly ($p < 0.01$) acceptable color (Eakes and Blumer, 1975b). However, DuBose et al. (1981) contended that cured, smoked hams prepared with 25, 75 or 156 mg/kg nitrite were not significantly ($p < 0.05$) different in color. Brine-cured, smoked turkey drumsticks treated with nitrite were found to be more acceptable (colorwise) than their nonnitrite treated counterparts (Olson et al., 1979). Brown et al. (1974) reported a difference in the color intensity of brine-cured hams containing 91 or 182 mg/kg nitrite. Color was darker for the product containing the greater nitrite level.

In comparison to brine- or dry-cured products, comminuted meats require lower nitrite levels for color development because the chopping/emulsification process increases the available surface area and enhances the distribution of nitrite. Wasserman and Talley (1972) and Hustad et al. (1973) reported an unpleasant gray color in unsmoked frankfurters prepared without nitrite in the cure. Similar results were found in the sensory evaluation of salami sausage (Skjelkvale and Tjaberg, 1974) and thuringer sausage (Dethmers and Rock, 1975). Hustad et al. (1973) reported no significant ($p < 0.05$) difference in the color acceptability of frankfurters prepared with 50, 100 or 156 mg/kg nitrite, and that only 25-50 mg/kg nitrite was necessary for a stable color. As little as 40 mg/kg nitrite resulted in acceptable color in chicken frankfurters (Gray et al., 1979) and in turkey frankfurters (Sales et al., 1980), while 50 mg/kg nitrite was necessary in a beef-pork bologna product (Lin and Sebranek, 1979) and in thuringer sausage (Dethmers and Rock, 1975). In general, as the level of nitrite input increased, products exhibited greater color acceptability (Sebranek et al., 1977) and

provided Hunter color values of more redness and less yellowness (Sales et al., 1980).

2. Flavor

a. Cooked, uncured meat flavor: The elusive character and composition of meat flavor have been extensively researched during the past 20 years. Many advances have been made and it is believed that information on the compounds and reactions involved in the formation of satisfactory flavor could be utilized in a number of ways, including (1) assessment of the best conditions for storing and processing meat, (2) improving the flavor characteristics by closer attention to breeding, and (3) producing high quality extracts or synthesizing better meat flavors which in turn could be used to impart flavor characteristics to meat analogs or to meat which is flavor deficient (Gordon, 1972; Wasserman, 1979).

Meat flavor can be considered to consist of four components, the volatile and nonvolatile fractions from both raw and cooked meat. In raw meat, the nonvolatile and volatile components are associated with taste and aroma, respectively, and include precursors of the cooked meat flavor. Therefore, upon heating, both components of raw flavor may form volatile and nonvolatile compounds which contribute to the cooked flavor of meat (Landmann and Batzer, 1966; Dwivedi, 1975).

The chemical basis of the nonvolatile (or precursor) and volatile fractions in a variety of meats has been extensively researched and reviewed. Results indicate that the nonvolatile precursors which are water soluble as well as fat soluble, are low molecular weight compounds and include glycoproteins, mononucleotides, reducing sugars, amino acids

and their degradation products (Batzer et al., 1960; Landmann and Batzer, 1966; Sanderson et al., 1966; Dwivedi, 1975). The volatile components of meat flavor, many of which are produced from the nonvolatile compounds during cooking, have been isolated and identified using gas chromatography and/or mass spectrometry (Bender and Ballance, 1961; Sanderson et al., 1966; Dimick et al., 1972; Hirai et al., 1973; Persson et al., 1975; Persson and von Sydow, 1973; Shibamoto et al., 1981). Gordon (1972), Dwivedi (1975) and Wasserman (1979) summarized the various volatile compounds isolated from beef, pork, lamb and poultry. They suggested that the relative concentration of all the components present determine the characteristic flavor of meat. Conversely, Chang and Peterson (1977) concluded that certain compounds, such as aliphatic and aromatic hydrocarbons, saturated alcohols, carboxylic acids, esters, ethers and carbonyl compounds (aldehydes and ketones) may not be primary contributors to meat flavor. However, they suggested that lactones, acyclic sulfur-containing compounds (mercaptans and sulfides), nonaromatic heterocyclic compounds containing sulfur, nitrogen and oxygen (hydrofuroanoids) and aromatic heterocyclic compounds containing sulfur, nitrogen and oxygen (pyrazines and thiophenes) are probably the main or most important contributors to meat flavor.

Different sources/types of meat have their individual and distinctive flavor and aroma. Most investigators generally agree that the fundamental meaty flavor is associated with the nonvolatile, water soluble components of the meat, while the species specific flavor is apparently associated with the volatile compounds which arise mainly from the fat during cooking (Landmann and Batzer, 1966). Chang and Peterson (1977)

suggested that fat may serve as a reservoir for cooked meat flavor, i.e., flavor precursors, which may be unique to a given type of meat are leached out into and stored in the fat. This is supported by the fact that refined, cooked animal fat on its own does not produce characteristic meaty flavors and aromas. However, lipids themselves are not responsible for the formation of the sulfur and nitrogen-containing heterocyclic compounds present in the volatiles of cooked meat (Chang and Peterson, 1977).

Despite the large number of compounds isolated from meat products, the components primarily responsible for the various characteristic flavors have not been identified (Chang and Peterson, 1977). In addition, the numerous factors which influence flavor and off-flavor in muscle foods must be considered. Sink (1979) reviewed the genetic (species, breed and sex), environmental (age, nutrition and stress) and processing (carcass washing, freezing, formulations, heating and smoking) factors that could affect flavor and aroma. A review on similar factors relative to off-flavor production has also been published (Reineccius, 1979). Both authors concluded that no single one group of factors can be assigned the role of "principal influencer", but certain aspects such as species and diet exert more pronounced effects than others. In their study of the influence of processing procedures on meat flavor, Landmann and Batzer (1966) concluded that canning, irradiation and freeze-drying alter either the qualitative or quantitative composition of the flavor system by the addition of extraneous chemical substances, by unavoidable chemical changes which are the direct result of processing, or simply by the loss of compounds responsible for flavor production.

b. Cured meat flavor: The close association between nitrite and cured meat flavor has been extensively researched and reviewed (Bailey and Swain, 1973; MacDougall et al., 1975; Gray et al., 1981b), but the chemical basis for this characteristic flavor is still not fully elucidated (Gray et al., 1981b). For the most part, the flavor volatiles isolated from dry-cured (Ockermann et al., 1964; Lillard and Ayres, 1969) and stitch-pumped (Cross and Ziegler, 1965; Piotrowski et al., 1970) hams were qualitatively similar to their uncured (nonnitrite-treated) counterparts, but there were quantitative differences. Bailey and Swain (1973) advocated that the accumulation of lipid oxidation end-products (e.g., carbonyls) in uncured, cooked meats created the flavor difference relative to cured products. Therefore, they concluded that a major contribution by nitrite is its activity in retarding the oxidation of lipids in cured, cooked meats. However, MacDougall et al. (1975) contends that since the nitrite ion is a reactive species, its reaction with other meat components could result in hitherto unidentified products. Such compounds together with many individual aromatic compounds could then produce the composite sensation referred to as "cured meat flavor".

The quantitative relationship between nitrite and cured flavor has been a matter of some debate. A common opinion is that, analogous to color formation, an adequate cured flavor is obtained with a relatively low nitrite concentration, an increase in nitrite producing no improvement in quality (MacDougall et al., 1975). However, the precise minimum level of nitrite to provide an organoleptically acceptable cured flavor in a product is influenced by many factors, including: (1) the type of meat used; (2) formulation of the product as to the

incorporation of spices; (3) processing procedure used--tumbled, stitch-pumped or brine-cured; (4) use and/or extent of smoking; (5) cooking tradition, such as how the product is prepared, duration of frying, level of fat desired; (6) effect of salt on increasing cured flavor perception (MacDougall et al., 1975); and (7) influence of sensory procedures on panelist's judgment; such as, sample appearance, score sheets and copresence of oxidized/rancid flavor (Price and Greene, 1978). These factors will be taken into account in the following review of the nitrite-cured meat flavor relationship found in bacon, comminuted products and cured hams.

Brooks et al. (1940), in a study of Wiltshire bacon, first described the relationship between nitrite and cured meat flavor. They advocated that as little as 10 mg/kg nitrite resulted in a satisfactory flavor. MacDougall et al. (1975) evaluated the same type of product and suggested that 100-150 mg/kg nitrite (i.e., 1500 mg/kg in the brine) was required for maximum flavor. Although MacDougall et al. (1975) did not conclude the minimum nitrite level necessary for organoleptically satisfactory bacon flavor, they did note that this research established that a flavor-producing reaction between meat and nitrite is continuing well-beyond the levels where color formation is completed. The influence of salt on bacon flavor was reported by Kimoto et al. (1976). They found that differences in bacon flavor were due to the presence of salt and not so much associated with the addition of nitrite. As a follow-up, bacon prepared with and without nitrite was evaluated by consumer preference (Wasserman et al., 1977) and according to the mean percentage plate waste, i.e., the amount of the sample left on a plate when served in an

institutional setting (Williams and Greene, 1979). Such research revealed that an identifiable cured bacon flavor can be produced without the use of nitrite, providing salt is incorporated in the formulation.

Comminuted products, such as beef-pork frankfurters (Wasserman and Talley, 1972; Hustad et al., 1973), chicken frankfurters (Gray et al., 1979), turkey frankfurters (Sales et al., 1980) and comminuted pork (Hadden et al., 1975) have been prepared with various levels of nitrite and reported to exhibit significantly ($p < 0.01$) more cured flavor than their nonnitrite-treated counterparts. The minimum level of nitrite necessary varied with the product. Sebranek et al. (1977) found that the flavor acceptance of beef-pork frankfurters increased with nitrite concentration (up to 156 mg/kg). At least 50 mg/kg was necessary for satisfactory flavor in thuringer sausage (Dethmers and Rock, 1975), and levels of 100 mg/kg nitrite or more were preferred. Conversely, Sales et al. (1980) reported that turkey frankfurters prepared with 40 mg/kg nitrite were not significantly ($p < 0.05$) different from samples with 100 mg/kg nitrite. For comminuted pork, Hadden et al. (1975) found that the incorporation of 156 or 200 mg/kg nitrite was preferred to the 20 mg/kg level. The type of meat used in the manufacture of frankfurters can be influential, as demonstrated in the study by Simon et al. (1973). For frankfurters containing beef and pork (50:50), no flavor difference was found between 39 and 78 mg/kg nitrite or between 78 and 156 mg/kg nitrite; however, the frankfurters prepared with 100% beef were acceptable to all nitrite levels and even without nitrite addition. Wasserman and Talley (1972) demonstrated the effect of smoke on cured frankfurter flavor. They reported that beef-pork samples prepared with 78 and 156

mg/kg nitrite were not significantly ($p < 0.01$) different in flavor acceptability when no smoke was used during the cooking. However, all smoked frankfurters were judged similar in flavor, regardless of whether or not sodium nitrite was used in their preparation. Price and Greene (1978) suggested that salt was the major contributor to the cured meat flavor of ground pork since nitrite alone at a level of 200 mg/kg did not produce much flavor. The combination of salt and nitrite provided the greatest cured flavor, but they advocated that, should nitrite use be prohibited, the desirable/characteristic flavor of comminuted products could still be obtained by incorporation of salt in the formulation.

Dry-cured hams (Brown et al., 1974; Kemp et al., 1974; Eakes and Blumer, 1975a,b; Kemp et al., 1975) and pork loins (Cho and Bratzler, 1970; Eakes and Blumer, 1975b) prepared with nitrite and/or nitrate exhibited greater cured flavor than their nonnitrite counterparts, i.e., controls with salt and sugar. Cho and Bratzler (1970) further noted that, in pork loins, this trend continued even when smoke was introduced or salt was omitted. However, Swain (1972) reported that their taste panel selected ($p < 0.001$) smoked and nitrite-treated hams as having better cured flavor than the nonsmoked, nonnitrited samples. When the same panel rated flavor desirability on a hedonic scale, smoked-nitrite hams were rated higher than nonsmoked-nitrite samples, and nitrite samples rated higher than nonnitrite samples. The precise nitrite level required for satisfactory flavor in cured hams varies. Brown et al. (1974) reported that hams cured with 91 mg/kg nitrite provided desirable flavor and that no improvement was noted when the nitrite level was increased to 182 mg/kg. MacDonald et al. (1980c) observed that the

introduction of 50 mg/kg nitrite into ham resulted in significant cured flavor development, while DuBose et al. (1981) found no significant ($p < 0.01$) flavor differences in hams cured with 25, 75 or 156 mg/kg nitrite.

For the most part, research indicates that both bacon and comminuted products with acceptable cured flavor could be manufactured with little or no nitrite as long as salt was included in the formulation. However, hams would still require at least 25-50 mg/kg of sodium nitrite.

c. Warmed-over flavor: The term, warmed-over flavor (WOF), was first coined by Tims and Watts (1958) to describe the rancid or stale flavor which develops within 48 hours in cooked meat held at 4°C. This flavor defect is even more apparent upon rewarming the product (Sato and Hegarty, 1971). The subject of WOF in meat, poultry and fish has been extensively reviewed by Pearson et al. (1977). They emphasized that the problem of WOF has become even more important as precooked meats have assumed an increasingly larger proportion of the market, due to the rapid growth of fast-food service facilities.

Since increased levels of various carbonyl compounds (pentanal, hexanal, n-nona-3,6-dienal) have been found to correlate with greater WOF, it has been hypothesized that warmed-over flavor is due to lipid oxidation (Tims and Watts, 1958; Kemp, 1974; Pearson et al., 1977). Based on this premise, most studies of WOF utilize two primary methods of analysis--the thiobarbituric acid (TBA) values for lipid oxidation and/or organoleptic evaluation for rancid flavors and odors.

Meat lipids are commonly classified as depot or adipose tissue and intramuscular or tissue lipids (Pearson et al., 1977). Although

adipose tissue consists mainly of triglycerides, the intramuscular lipids contain triglycerides, phospholipids and lipoproteins (Love and Pearson, 1971; Pearson et al., 1977). Phospholipids due to their high unsaturated fatty acid content, tend to undergo rapid oxidation and are at least partially responsible for the off-flavors which develop during storage of cooked, uncured meat (Younathan and Watts, 1959; Bailey and Swain, 1973; Love and Pearson, 1971, 1976; Fooladi et al., 1979). However, other factors such as cooking (Igene et al., 1979), chopping/emulsification (Sato and Hegarty, 1971), species (Wilson et al., 1976), and the relative concentrations of red and white fibers (Wilson et al., 1976) must be considered relative to acceleration of oxidation.

For many years, heme proteins were implicated as the major prooxidants of lipid oxidation in meat products (Younathan and Watts, 1959; Zipser et al., 1964). However, after Eriksson et al. (1971) demonstrated that acid- or heat-denatured heme proteins caused greater nonenzymatic lipid oxidation due to increased exposure of the heme group, research examined the influence of nonheme iron on meat lipids. Love and Pearson (1974) and Sato and Hegarty (1971) initially contended that nonheme iron acted as a prooxidant in cooked meat, while heme protein iron had little or no effect. However, later research indicated that both iron sources could act as prooxidants depending upon the conditions (Love and Pearson, 1976). Heme proteins are most active catalysts of lipid oxidation when the iron is in the ferric (Fe III) state, while nonheme iron is a more active catalyst in the ferrous (Fe II) state (Greene and Price, 1975). Ascorbic acid aids the nonheme iron catalysis by maintaining it in the ferrous state (Sato and Hegarty, 1971). Research by Igene et al. (1979)

established that removal of heme pigment from cooked meat inhibited lipid oxidation according to chemical and sensory analyses. However, they also determined that cooking released a significant amount of nonheme iron from bound heme pigments, which accelerated lipid oxidation in cooked meat. Therefore, it was concluded that the increased rate of lipid oxidation in cooked meat was due to release of nonheme iron during cooking (Igene et al., 1979). In addition, to the heme and nonheme iron, salt is believed to catalyze lipid oxidation, although the mechanism is not known (Love and Pearson, 1971).

Nitrite is believed to have an antioxidant role since it has been shown to retard lipid oxidation or the development of WOF in cooked meat and in processed meat products. The correlation between nitrite addition and reduced TBA values and/or less detectable off-flavors/off-odors has been indicated in studies on ham (Swain, 1972; Price and Greene, 1978; MacDonald et al., 1980b,c), turkey frankfurters (Sales et al., 1980), chicken frankfurters (Gray et al., 1979), preblended sausage and frankfurters (Waldman et al., 1974), comminuted pork (Hadden et al., 1975), thuringer sausage (Dethmers and Rock, 1975), bologna (Lin and Sebranek, 1979) and in nitrite-treated beef, pork or chicken meat systems (Fooladi et al., 1979; Igene et al., 1979). However, the actual mechanism by which nitrite minimizes WOF is not fully understood. Current hypotheses include: (1) nitrite may stabilize the lipid components of the membranes (Pearson et al., 1977); such as, protecting against the oxidation of phospholipids (Fooladi et al., 1979); (2) nitrite may inhibit the natural prooxidants in the muscle (Pearson et al., 1977); such as, chelating trace metals (MacDonald et al., 1980a); or

(3) nitrite may react with meat components to form compounds with antioxidant properties (Kanner, 1979a,b).

3. Antibotulinal Activity

The ability of sodium nitrite to inhibit the growth and toxin production of C. botulinum is undoubtedly its most important function. The broad research areas of botulism and the antimicrobial efficacy of nitrite in cured meats has been extensively reviewed by Sofos et al. (1979a). The reader is directed to this publication for more details on the subject which will be briefly summarized.

Since soil is the primary ecological niche of C. botulinum, all food must be considered to be contaminated with spores of the organism, either by direct contact with the soil or indirectly via airborn dust (Lechowich et al., 1978). Although seven types (A through G) of C. botulinum have been isolated, types A and B are the major causes of botulism in heat processed foods, because the high heat resistence of their spores permits survival in under-processed foods (Sofos et al., 1979a).

C. botulinum can be inhibited by storage of food below 3°C; 10% NaCl (brine concentration); a pH value below 4.5 or by a proper combination of these factors (Riemann et al., 1972). Most meat products do not have the pH value or brine concentration required to completely inhibit C. botulinum, and there is always a risk of temperature abuse (Riemann et al., 1972). Despite these adversities, the botulinal safety of cured meat products is well documented (Lechowich et al., 1978; Tompkin, 1980). As indicated in Table 1 (Sofos et al., 1979a), only 5%

of all botulism outbreaks that occurred in the United States during the period of 1899-1973 were due to meats, while vegetables, fish, fruit and condiments accounted for over 80% of the incidences. Although many factors contribute to this excellent public health record, the use of nitrite is considered to be the primary reason. Indeed, there are regular outbreaks of botulism from home-cured meats in France and Spain where nitrite and/or nitrate are not used or used under poorly controlled conditions (Roberts, 1975). In contrast, no botulism outbreaks have resulted from commercially cured meats in countries where nitrite is commonly used, despite the demonstratable presence of C. botulinum (Roberts, 1975).

Table 1. Botulism outbreaks[1]

| Food Processing Type | Period 1899-1949 | 1950-1973 | 1899-1973 | % Total |
|---|---|---|---|---|
| Home processed | 382 | 113 | 495 | 72.0 |
| Commercially processed | 48 | 14 | 62 | 9.0 |
| Unknown | 47 | 84 | 131 | 19.0 |
| **Food Product** | | | | |
| Vegetables | -- | -- | 150 | 68.5 |
| Fish and Fish Products | -- | -- | 29 | 13.2 |
| Condiments | -- | -- | 20 | 9.2 |
| Meats | -- | -- | 11 | 5.0 |
| Other | -- | -- | 9 | 4.1 |

[1]Source: V. G. Bowen, J. G. Cerveny and R. H. Deibel. Effect of sodium ascorbate and sodium nitrite on toxin formation of Clostridium botulinum in weiners. Applied Microbiol. 27:605, 1974.

Like most of the functions of nitrite, the precise mechanism(s) through which nitrite inhibits C. botulinum growth and toxin production is still not known. Johnston et al. (1969) hypothesized that some possible roles of nitrite in maintaining meat stability are: (a) to enhance the destruction of spores by heat, (b) to increase the rate of spore germination during thermal processing, with subsequent killing of germinated spores by heat, (c) to prevent growth of the germinated spores which survive thermal processing, or (d) to react with some component(s) of the meat to form an antimicrobial compound(s). Subsequent research has eliminated most of these suggestions, either because the nitrite level necessary for a particular effect was 6-10 times more than that used in normal meat curing (Duncan and Foster, 1968) and/or that mechanisms demonstrated in model systems were not also found in actual meat products (Ashworth and Spencer, 1972; Sofos et al., 1979a). However, the influence of nitrite in preventing outgrowth of the germinated spores is still a viable explanation. Using a Liver Veal Agar medium, Duncan and Foster (1968) demonstrated that 0.01% sodium nitrite at pH 6.0 prevented cell division of vegetative cells, causing them to lyse. Tompkin et al. (1978c) suggested that nitric oxide (formed from residual nitrite via nitrous acid) reacts with iron in the vegetative cells, thereby blocking some metabolic step essential for outgrowth. This reaction might involve the iron in ferredoxin or an enzyme in which iron plays an essential role (Tompkin et al., 1978c). Yarbrough et al. (1980) agreed with this theory, but advocated two others. First, nitrite interferes with energy conservation by inhibiting oxygen uptake, oxidative phosphorylation

and proton-dependent active transport, and secondly, nitrite acts as an uncoupler, causing collapse of the proton gradient.

After reviewing the conflicting research pertaining to the antibotulinal mechanism of nitrite, Sofos et al. (1979a) contended that no single mechanism seems to entirely explain the nitrite effect on the safety of cured meat products, and to apply in culture media and all types of meat products. They concluded that most of the results are true and important for the conditions and systems tested in each particular case, and that the effectiveness of nitrite in delaying botulinal toxicity is probably due to one or more mechanisms, or one or more factors involved in each particular product or system studied.

It is a well-known fact that a majority of the 156 mg/kg sodium nitrite added to meat products is used for the control of C. botulinum and only a small fraction (25 mg/kg) is needed for the development of the characteristic color and flavor of the products (Sofos et al., 1979a). The precise minimum level of nitrite necessary to insure the microbial safety of meat products is controversial. Bowen et al. (1974) and Hustad et al. (1973) contended that as low as 50 mg/kg nitrite inhibited toxin formation in frankfurters for 56 days at 27°C. However, many other authors believe that at least 100-200 mg/kg nitrite is required to inhibit C. botulinum under the environmental conditions found most meat products (Christiansen et al., 1973, 1974, 1975; Collins-Thompson et al., 1974; Tompkin et al., 1977). Factors that aid the antibotulinal efficacy of nitrite, and thereby reduce the level of nitrite required, must be considered. These include, acidic conditions (Christiansen et al., 1975; Wasserman and Fiddler, 1976; Christiansen,

1980), refrigerator storage conditions (Collins-Thompson et al., 1974), low innoculum levels (Hustad et al., 1973; Christiansen et al., 1974; Collins-Thompson et al., 1974), and reduced levels of available iron (Tompkin et al., 1978c,e, 1979a).

Aside from the quantitative relationship between nitrite and botulinal inhibition, the major issue is the relative importance of initial nitrite input versus that of the residual nitrite found in the product after processing and during storage. For several years, researchers contended that the input level of nitrite was the important factor. Studies with inoculated meat systems (Hustad et al., 1973; Christiansen et al., 1974, 1975; Sofos and Busta, 1980) have demonstrated that increased levels of formulated nitrite decreased the probability of botulinal toxin production. Tompkin et al. (1977) established a base line for inhibition of C. cotulinum by nitrite in a canned meat product, and predicted the average time to first swell to be 6.7, 29.8, 82.6 and 94.3 days when 0, 50, 100 and 156 mg/kg nitrite was added to the meat. Using the same type of product, Christiansen et al. (1973) pointed out that two different input levels of nitrite (at 50 and 156 mg/kg) both resulted in a residual nitrite content of 5 mg/kg within 21 days at 27°C, but the degree of inhibition was much greater for the higher initial level of nitrite.

However, these authors failed to observe that as the nitrite levels decreased, so did the C. botulinum cell level (Christiansen, 1980). Therefore, Christiansen et al. (1978) revised their theory and advocated that their data indicated a race between nitrite depletion and death of the germinated botulinal spores. The safety of meat products

is dependent upon sufficient residual nitrite until the viable cell level has decreased to a point at which growth can no longer be initiated (Christiansen et al., 1978).

The idea that residual nitrite levels are important is enhanced by results of experiments in which nitrite is depleted prior to temperature abuse or nitrite is depleted rapidly during abuse (Tompkin et al., 1978c; Christiansen, 1980). For example, Tompkin et al. (1978b) held inoculated, canned cured meat in a refrigerator for 0-26 weeks prior to temperature abuse at 27°C. As the refrigerator (4.4°C) storage time increased, the germinated cell count remained stable, but the residual nitrite and resultant degree of inhibition both decreased. Christiansen (1980) suggested that this type of experiment be done to evaluate any future changes in nitrite usage as it simulates practical conditions, i.e., temperature abuse of the product occurring after an extended refrigerator storage.

As previously mentioned, many factors other than the presense of nitrite contribute to the impressive botulinal safety record of meats. Despite the ubiquitous nature of C. botulinum spores (Lechowich et al., 1978), raw meat and poultry products exhibit a low incidence of contamination, reportedly only 1-7 spores/pound of meat (Holley, 1978; Lechowich et al., 1978; Sofos et al., 1979a; Tompkin, 1980). This, by itself, would appear to explain the infrequent incidence of botulism in meat. However, in consideration of the tonnage of meat which is produced and consumed, the potential of botulism is quite apparent (Lechowich et al., 1978) and other factors must be considered. Most authors contend that inhibition is due to the interacting effects of several factors;

such as, salt concentration, water activity, pH value, heat treatment, nitrite concentration, $E_h$, product composition, packaging, storage conditions, initial contamination, inoculum level, presence of iron and the level of reductants and/or chelators in the product (Christiansen et al., 1973, 1974, 1975; Hustad et al., 1973; Collins-Thompson et al., 1974; Roberts, 1975; Lechowich et al., 1978; Lee et al., 1978; Sofos et al., 1979a; Cerveny, 1980; Christiansen, 1980).

B. NITRITE AND N-NITROSAMINE FORMATION

Although nitrite contributes to the color, flavor and botulinal safety of cured meat products, its continued use is questionable. It is generally known that nitrite (as nitrous acid) will interact with the primary, secondary or tertiary amines, polyamines and quaternary ammonium compounds found in many food systems, to form N-nitrosamines. Over 25 years ago, it was reported that severe liver damage (Barnes and Magee, 1954) and the induction of liver tumors (Magee and Barnes, 1956) occurred in rats after administration of N-dimethylnitrosamine (NDMA). This was the first report that N-nitrosamines were carcinogenic, but was subsequently followed by evidence of the mutagenic, teratogenic and embryopathic properties of many N-nitroso compounds (Shank and Newberne, 1976). Recent research has even questioned the possible carcinogenic effects of nitrite itself (Newberne, 1979), but a follow-up report indicated that Newberne's interpretation of the data was suspect (Ember, 1980). Although there is no direct evidence that N-nitroso compounds are carcinogenic to man, indirect proof from animal studies on various species would suggest the potential danger to man (Gray and

Randall, 1979). Many reviews on N-nitrosamines have been published in the past few years, dealing with their formation and occurrence in foods and their toxicology and human health hazards (Sebranek and Cassens, 1973; Foreman and Goodhead, 1975; Scanlan, 1975; Fishbein, 1979; Gray and Randall, 1979).

As discussed by Scanlan (1975) and Foreman and Goodhead (1975), the overall kinetics of N-nitrosamine formation are influenced by nitrite concentration, pH, temperature and type/concentration of the amine. They summarized research which demonstrated that the rate of N-nitrosamine formation is directly proportional to the amine concentration and to the square of the nitrite concentration. Since the interaction is between the undissociated nitrous acid and an unprotonated amine, then the reaction rate is maximized at pH 3.4, increases as the basicity of the amine decreases and doubles with each 10°C increase in temperature above ambient (Scanlan, 1975; Foreman and Goodhead, 1975).

In surveys of the volatile N-nitrosamines isolated from various food systems, cured meats are of greatest concern since nitrite is directly added during processing. However, the cured meat group is divided into two factions on the basis of N-nitrosamine occurrence. One group includes bacon, where N-nitrosamines have been consistently found, and the other group includes hams, frankfurters, bologna and similar products in which N-nitrosamines have only been isolated sporadically (Sofos et al., 1979a).

NPYR and NDMA are the volatile N-nitrosamines found most commonly and in the greatest quantity in fried bacon. NDMA is usually reported in the range of 1-5 μg/kg for the total fried bacon samples (Gough, 1977; Sen et al., 1979). Sometimes NDMA is not detected at all, but there are

also isolated cases of higher concentrations, such as the 30 µg/kg NDMA reported by Sen et al. (1973). In contrast, NPYR is consistently found in fried bacon. The concentration may range from 5 to 25 µg/kg (Sen et al., 1973; Gough, 1977), but has also been reported at levels up to 139 µg/kg (Harvey et al., 1976) or 320 µg/kg (Nitrite Safety Council, 1980).

For other cured meats, N-nitrosamines have been sporadically isolated and then at levels below 25 µg/kg. Several surveys have reported no detectable (<1 µg/kg) volatile N-nitrosamines in thuringer sausage (Dethmers and Rock, 1975), baby food containing cured meat (Sen et al., 1973, 1974; Harvey et al., 1976), frankfurters (Fiddler et al., 1972; Hustad et al., 1973; Nitrite Safety Council, 1980), canned comminuted ham (Christiansen et al., 1973), bologna (Nitrite Safety Council, 1980) and salami (Nitrite Safety Council, 1980). Some studies have found volatile N-nitrosamines in such products, but at levels below 10 µg/kg (Gough, 1977; Sen et al., 1979; Evans-Holland, 1980; Nitrite Safety Council, 1980; Gray et al., 1981a).

The vast differential in N-nitrosamine levels found in bacon and other cured meat products led researchers to analyze conditions which influence nitrosamine (particularly NPYR) formation in bacon. Three primary factors were investigated including, cooking conditions such as the time-temperature relationship, role of adipose and lean composition, and the nitrite concentration of the product. Very early in the N-nitrosamine analyses it was observed that no N-nitrosamines are found in the raw sample, but detectable levels of NPYR are produced upon frying (Sen et al., 1973; Pensabene et al., 1974; Harvey et al., 1976;

Sen et al., 1979). Pensabene et al. (1974) studied the effect of frying and cooking conditions on NPYR formation in bacon and concluded that N-nitrosamine formation is primarily dependent on frying temperature, but not time. Samples from one belly formed no NPYR when fried for 1-5 minutes at 99°C, while samples from the same belly, fried to the same "doneness" at 204°C for 4 minutes produced 17 μg/kg of NPYR. These authors also compared pan-frying to other cooking methods and observed that standard frying procedures produced high yields of NPYR (5-20 μg/kg); no detectable levels were isolated when the product was microwaved, while baking or broiling produced variable amounts.

Subsequent research on bacon revealed that higher N-nitrosamine levels were found in the adipose and/or cooked-out fat than in the lean portion of bacon (Havery et al., 1976; Gray and Collins, 1978; Bharucha et al., 1979; Sen et al., 1979). Initially, it was theorized that a time-temperature relationship was the major determinant of this distribution. Fat, due to its lower moisture content, would reach a higher temperature in a shorter time than the lean, therefore higher N-nitrosamine levels (especially NPYR) would result (Coleman, 1978). However, Fiddler et al. (1974) subsequently demonstrated that lean, when separated from the adipose in bacon, does not contain NPYR when uncooked, fried alone or fried with Crisco. In contrast, adipose tissue which was isolated from the same bacon sample, contained no NPYR when uncooked, but high concentrations were detected upon frying (Fiddler et al., 1974). They hypothesized that the level of collagen or connective tissue in the adipose of bacon was an influential factor in these results because such tissues contain high levels of proline and hydroxy proline, believed to

be possible NPYR precursors (Gray and Dugan, 1975; Bharucha et al., 1979). This theory was substantiated by research on the morphology of bacon by Cassens et al. (1979b). These investigators reported that the adipose of bacon is composed of lipid-filled cells or adipocytes, which are each surrounded by cytoplasmic protein. Nitrite-containing brines go to the surface of fat globules via channels of the extracellular space, capillaries, connective tissue and layers of cytoplasmic protein. This close interface between protein and nitrite presents a unique site for reaction and increases the chance of NPYR formation (Cassens et al., 1979b). NPYR, being very fat soluble, will then partition into the fat globule (Fiddler et al., 1974).

In similar studies on the distribution of NPYR in bacon, it was observed that only a portion of the total N-nitrosamines produced upon frying the sample are actually isolated in the adipose and/or lean. Research revealed that 25-50% of the total NPYR (Gray and Collins, 1977; Gray et al., 1978; Bharucha et al., 1979) and 60-80% of the total NDMA (Gray et al., 1978; Bharucha et al., 1979) are found in the vapor phase during the cooking procedure.

As previously mentioned, N-nitrosamine formation is proportional to the square of the nitrite concentration. This fact has been verified in fried bacon (Sen et al., 1974; Gray and Collins, 1978; Pensabene et al., 1979). Pensabene et al. (1979) reported that the NPYR and NDMA levels in the edible portion, drippings and total sample of fried bacon were highly correlated ($p < 0.01$) with residual and added nitrite content. This contrasts with the findings of Sen et al. (1974) who stated that the NPYR found in fried bacon correlated ($p < 0.001$) with the initial level

of nitrite added, but not with the residual levels found just prior to frying. They concluded that an intermediate (or precursor) N-nitroso compound was produced in the early stages of curing and its concentration was dependent upon the initial nitrite concentration.

In comparison to bacon, other cured meat products have exhibited different results when exposed to similar conditions. The cooking of frankfurters, bologna or hams by frying, boiling or broiling has not resulted in increased N-nitrosamine formation (Hustad et al., 1973; Sen et al., 1979; Nitrite Safety Council, 1980), nor does increased nitrite input result in increased N-nitrosamine levels (Christiansen et al., 1973; Hustad et al., 1973; Dethmers and Rock, 1975; Nitrite Safety Council, 1980). In fact, Fiddler et al. (1972) demonstrated that ten times the legal limit of sodium nitrite must be added to frankfurter formulation before NDMA levels exceeding 10 μg/kg can be detected. In contrast, Sen et al. (1974) observed that for fried bacon 50-100 mg/kg nitrite resulted in 2-10 μg/kg levels of nitrosamines. The negligible levels of volatile N-nitrosamines found in cured meats other than bacon could be attributed to the greater amount of adipose tissue in bacon compared to lean meats (Cassens et al., 1979b). As previously discussed, the interface between nitrite and collagen at the fat globule surface is important in nitrosamine formation in bacon. In highly comminuted products (frankfurters, bologna, sausages) the adipose tissue is largely destroyed, and the lipid is released being redispersed in the co-called "emulsion" form (Cassens et al., 1979b).

Commercially cured products (other than bacon) will sporadically contain higher levels of N-nitrosamines and no adequate explanation is

available. Fiddler et al. (1972) suggested a number of variables including, localized high concentrations of nitrite in emulsions due to inadequate homogenization during processing, age and condition of the meat, nitrite concentration, the type and amounts of other ingredients used, the actual processing conditions, and the subsequent time and temperature of storage. At one time, the spice-cure mixes added to these products were suspected of contributing N-nitrosamines. Spices and curing salts were originally packaged together, which allowed amines and nitrite to interact. NDMA, NPYR and N-nitrosopiperidine (NPIP) were detected in such mixtures at levels of 50-2000 μg/kg (Havery et al., 1976). By 1974, federal laws required spice-cure mixes to be packaged "piggy-back", i.e., separate packaging of the spices and curing salts, and this source of volatile N-nitrosamines in such products as frankfurters, bologna and sausages has been eliminated (Havery et al., 1976).

The amine precursors of the most consistently reported volatile N-nitrosamines, NPYR and NDMA, have been thoroughly researched. Although N-nitrosoproline, proline, collagen, putrescine, spermidine and pyrrolidine are believed to be potential precursors of NPYR in cooked bacon (Bills et al., 1973; Gray and Dugan, 1975; Gray and Collins, 1977, 1978), proline appears to be the most probable choice. The mechanism for NPYR from proline is not fully elucidated, but two theories have been proposed. Proline could be decarboxylated to form pyrrollidine, with subsequent formation of the N-nitroso derivative (Ender and Ceh, 1971), or proline could be N-nitrosated and subsequently decarboxylated to NPYR (Lijinsky and Epstein, 1970; Bharucha et al., 1979). The latter sequence appears to be more likely (Bharucha et al., 1979), but Nakamura et al.

(1976) emphasized that either mechanism is dependent on the cooking temperature.

Much less research has been conducted on the precursors of NDMA. Sarcosine and lecithin have been demonstrated to contribute to NDMA formation during the frying of bacon, but the principal precursor remains to be determined (Gray et al., 1978).

## C. NITRITE SUBSTITUTES

Due to the involvement of nitrite in the formation of N-nitrosamines in cured meat products, the Secretary of Agriculture established an Expert Panel on Nitrites, Nitrates and Nitrosamines in 1973 to examine the role of nitrite and nitrate in cured meats and their public health significance as related to botulism and N-nitrosamines (Sofos et al., 1979a). Based on data from the industry and other institutions, the Panel issued its final report in February, 1978 with the following recommendations (Anon., 1978a): (a) use of nitrate be discontinued in all meat and poultry products, except dry-cured products and fermented sausages, (b) the nitrite level permitted for curing of meat be limited to 156 mg/kg in all cured meat products, except bacon and dry-cured/fermented sausages which would be limited to 120 and 100 mg/kg, respectively, (c) the permitted residual nitrite level should be reduced from 200 to 100 mg/kg in cooked sausage products, 125 mg/kg in canned and pickle-cured products, 80 mg/kg in bacon and 50 mg/kg in canned, cured sterile products, and (d) alternative preservatives with the potential to replace or reduce nitrite in cured meats should be evaluated.

As reviewed by Sebranek (1979), initial research for potential nitrite alternatives was based on finding a substance or combination of

substances which could completely replace nitrite. Howard et al. (1973) evaluated 24 nitrogenous ligands, including pyridine, amino acids and amino acid esters, as to their ability to form ferrohemochromes with bovine myoglobin in model and meat systems. Methyl and hexyl nicotinate and N,N-diethylnicotinamide produced stable pink pigments in nitrite-free, cooked ground meat mixtures. When combined with 10-20 mg/kg nitrite, the pink color was even more stable and acceptable (Howard et al., 1973). However, reports have contended that the pigments produced by these compounds are easily oxidized and that nicotinic acid and its derivatives may have vasodilatory properties (Kemp, 1974; Dymicky et al., 1975). In a similar study of 300 nitrogenous compounds added to a meat slurry, Dymicky et al. (1975) reported that pyridine compounds (especially 3-acylpyridines) were the most effective pigment producers, depending on the nature and position of the substituent. The incorporation of betalain (beet) pigments (von Elbe et al., 1974) and/or other red food colors (Knowles et al., 1974) into nitrite-free sausages or luncheon meat is another alternative. MacNeil and Mast (1973) and MacNeil et al. (1973) investigated the use of spice extractives in frankfurters prepared without nitrite or nitrate. They reported that nitrite-free frankfurters of acceptable flavor and shelflife could be obtained when a spice extractive at 0.03 and 0.05% was added. However, a lack of the characteristic pink color in the product was a distinct disadvantage (MacNeil et al., 1973). As mentioned in the flavor section, the major contribution by nitrite to the characteristic flavor of cured meats is its activity in retarding the oxidation of lipids in such products (Bailey and Swain, 1973). If this is true, then antioxidants incorporated into cured meats

should be able to replace nitrite in flavor production. MacDonald et al. (1980b,c) investigated the effect of butylated hydroxytoluene (an antioxidant) and citric acid (a chelator) on the flavor, odor and lipid stability of nitrite-free ham. Although both of the additives were effective in reducing lipid oxidation and off-flavor development, they could not produce a typical cured ham aroma or flavor comparable to the nitrite-treated samples.

Obviously, there is not another compound or group of compounds which can emulate all the effects/functions of nitrite. Therefore, researchers altered their course of direction toward investigating substances which would block or inhibit N-nitrosamine formation when added to products prepared with reduced levels of nitrite. From their studies of N-nitrosamine formation, Bharucha et al. (1979) suggested that a good N-nitrosamine blocking agent should satisfy the following requirements: (1) serve as a good NO· radical trap (since NPYR formation is believed to be a radical mechanism), (2) be fat soluble (lipophilicity), (3) be non-steam volatile, and (4) be stable up to the maximum frying temperature of about 174°C.

Gray and Dugan (1975) tested the effect of several compounds on the N-nitrosation reaction in both aqueous and low moisture carboxymethylcellulose systems, and concluded that any compound which can react with nitrite can be utilized to at least partially inhibit the N-nitrosation reaction between a secondary amine and sodium nitrite. Compounds endogenous to meat, such as amino acids (cysteine, glutathione, methionine) as well as various substances added for preservative purposes (sodium bissulfite, tannic acid) have been found effective in inhibiting

N-nitrosamine formation (Gray and Dugan, 1975). Gray and Dugan (1975) and Coleman (1978) have reported that phenolic-type antioxidants, such as propyl gallate, tertiarybutyl hydroquinone, ethoxyquin and α-tocopherol can block N-nitrosamine formation. This related to the conclusion of Bharucha et al. (1979) that NPYR formation is by a radical mechanism. Substances added to the formulation of cured meats; such as, salt, nitrate and sodium acid pyrophosphate, do not appear to influence N-nitrosamine formation (Fiddler et al., 1973a,b), but others (glucono-delta-lactone and sodium tripolyphosphate) may have inhibitory properties (Fiddler et al., 1973a).

Two of the most thoroughly researched blocking agents are the reductants, ascorbate and its isomeric form, erythorbate. As discussed in the color section, these compounds are frequently incorporated in cured meat formulations to accelerate color development. Ascorbate and erythorbate are known to increase the depletion of residual nitrite in cured products (Brown et al., 1974; Sebranek et al., 1977), and therefore both compounds have been extensively investigated as potential inhibitors of N-nitrosamine formation. Studies of the reductants in buffer-model systems (Mirvish et al., 1972; Fan and Tannenbaum, 1973; Gray and Dugan, 1975; Mottram et al., 1975), bacon (Mottram et al., 1975) and frankfurters (Fiddler et al., 1973a,b) revealed that an ascorbate-nitrite ratio of 2:1 effectively inhibited (95-100%) the formation of NDMA (Fiddler et al., 1973b; Mottram et al., 1975), N-nitrosomorpholine (NMOR) (Mirvish et al., 1972; Fan and Tannenbaum, 1973) and N-nitrosopiperazine (Mirvish et al., 1972). Research has also concluded that ascorbate and erythorbate behave similarly and exhibit the same inhibitory activity (Fiddler et al.,

1973b). A similar conclusion was reached in comparing the blocking effects of the acid and salt forms of both reductants (Fiddler et al., 1973a).

It was observed that ascorbate does not degrade N-nitroso derivatives of amines, so initial theories on the mechanism of ascorbate proposed that nitrite and ascorbate must react in a manner that makes nitrite unavailable for N-nitrosation reactions with amines (Mirvish et al., 1972; Fan and Tannenbaum, 1973). This postulate has been substantiated by recent research (Williams, 1978; Fox et al., 1981) that nitrite (as nitrous acid) is bound to ascorbate in the form of various N-nitroso derivatives (3-nitrosoascorbate, 2,3-dinitrosoascorbate). The N-nitrosated ascorbate molecules can form dimers or degrade to various nitrogen oxides and dehydroascorbic acid. Further oxidation and N-nitrosation can follow to form ene-diol structures and nitroso-diketogulonic acid (Fox et al., 1981).

The relative effectiveness of ascorbate and erythorbate has three major limitations. First, the binding between nitrite and ascorbate (and resulting inhibitory effects) is pH sensitive (Fan and Tannenbaum, 1973; Mottram et al., 1975). The ascorbate anion ($pK_a$ 4.29) is nitrosated 240 times more rapidly than ascorbate, so the reductants are more effective at pH 3-5 (Mirvish et al., 1972). Second, the rate at which a given amine is N-nitrosated will determine the inhibitory efficacy of ascorbate. The faster an amine is N-nitrosated, the less effective ascorbate is at blocking the reaction because both the amine and the reductant would compete for the N-nitrosating agent (nitrous acid or nitrous anhydride) (Mirvish et al., 1972). Lastly, since ascorbate is

water soluble it has no effect on the nitrosation reactions in the fat/adipose tissue, the major site of nitrosamine formation in bacon (Mottram et al., 1975; Mottram and Patterson, 1977).

As a result of these findings, several fat soluble N-nitrosamine inhibitors have been studied. In both buffer-model systems (Mottram and Patterson, 1977) and commercial bacon (Sen et al., 1976; Bharucha et al., 1980), the incorporation of 500-1000 mg/kg of ascorbyl palmitate has resulted in reduced NPYR formation after cooking, while the use of sodium ascorbate was less effective and provided inconsistent results. Other lipophilic substances such as propyl gallate and piperazine hydrate were as effective as ascorbyl palmitate, and the authors concluded that all three compounds act by competing for nitrite with the various N-nitrosatable precursors of NPYR (Sen et al., 1976). Bharucha et al. (1980) observed that ascorbyl palmitate tends to lose activity on storage. They suggested the use of long-chain acetals of ascorbic and erythorbic acids, which inhibit N-nitrosamine formation by 93-98% and retain their efficacy in bacon even after 35 days storage at 3°C.

Another fat-soluble blocking agent which has received attention is α-tocopherol. Fiddler et al. (1978) injected pork bellies with a conventional cure formulation that would produce target levels of 125 mg/kg sodium nitrite and either 500 mg/kg sodium ascorbate or α-tocopherol, alone or in combination. A mixture of α-tocopherol and Polysorbate 20 (1:0.4 wt/wt) dispersed in the cure produced a good distribution of α-tocopherol in the adipose tissue (Fiddler et al., 1978). Both bacon (Fiddler et al., 1978) and model system (Pensabene et al., 1978) studies have revealed that 500 mg/kg α-tocopherol, alone or combined with sodium

ascorbate, inhibited NPYR formation more effectively than ascorbate alone.

Although many substances which can inhibit N-nitrosamine formation have been researched, investigators have also evaluated compounds that could block C. botulinum growth and toxin production, when added to meat products cured with reduced levels of nitrite. The effectiveness of ascorbate and/or erythorbate in controlling toxin formation was of concern because conceivably, ascorbate could enhance the growth of C. botulinum by decreasing the redox potential in the product or by lowering the residual nitrite concentration and thereby reducing the effectiveness of nitrite inhibition (Bowen et al., 1974). On the other hand, ascorbate could potentiate the inhibition by nitrite (Bowen et al., 1974). In their study on frankfurters, Bowen et al. (1974) reported that sodium ascorbate at levels of 105 and 655 mg/kg did not potentiate or decrease the inhibition of C. botulinum toxin formation by sodium nitrite. In contrast, Tompkin et al. (1978a) found that the incorporation of erythorbate alone (0.02%) in a canned comminuted pork system did not affect botulinal outgrowth, but the combination of 0.02% reductant with 50 mg/kg sodium nitrite was as effective as 156 mg/kg nitrite alone. In another study, Tompkin et al. (1978d) investigated the mechanism behind ascorbate potentiation of nitrite inhibition. Since BHT, TBHQ or sodium sulfide did not enhance the effect of nitrite, it was determined that neither the antioxidant or reducing properties of ascorbate/erythorbate were involved in the mechanism (Tompkin et al., 1978d). Rather, their data indicated that these compounds enhanced the effect of nitrite by sequestering a metal ion(s) in the meat. It is suggested that nitrite (nitric oxide)

reacts with a cation dependent material within the germinated botulinal cell and blocks a metabolic step which is essential for outgrowth (Tompkin et al., 1978d). Enhancement of nitrite by ascorbate/erythorbate may be due to preventing repair of damaged material or formation of new cation dependent material (Tompkin et al., 1978d).

Later, Tompkin et al. (1979b) qualified the antibotulinal effectiveness of ascorbate. Ascorbate enhanced inhibition when incorporated at levels below 200 mg/kg and/or when the product was temperature abused at the time of manufacture, but it decreased inhibition if used in excessive levels (>200 mg/kg) and/or when the product was stored prior to abuse. The first effect was due to sequestration of cations, while the second was because ascorbate hastened the depletion rate of residual nitrite (Tompkin et al., 1979b).

As mentioned in the antibotulinal section, the presence of significant levels of iron can inhibit the effectiveness of nitrite against C. botulinum. Tompkin et al. (1979a) reported that 500 mg/kg EDTA was not only more effective than 200 mg/kg ascorbate in sequestering iron, but there was also no evidence that EDTA hastens nitrite depletion as does ascorbate. Therefore, the authors proposed that a minimum of ascorbate/erythorbate be used in meats to hasten the curing reaction and stabilize color and flavor, but also supplement with a low level of EDTA for improved botulinal protection.

Since C. botulinum inhibition is pH dependent, researchers have studied the use of "acid development" to provide safe meat products without the use of nitrite. Riemann et al. (1972) suggested that 1% (or more) glucose should be added to a product because in the event of temperature

abuse, the glucose would be fermented to lactic acid by the indigenous microflora. As a result, the initial pH of the product would be reduced to a level at which the salt concentration is inhibitory (Riemann et al., 1972). The same authors stated that incorporation of sufficient glucono-delta-lactone would achieve the same effect. In a related study, Tanaka et al. (1980) reported that addition of Lactobacillus plantarum, a lactic acid former, along with a fermentable carbohydrate could lower the amount or completely eliminate sodium nitrite in bacon. Such preparations produce acid (causing a rapid decline in pH) only when the product is temperature abused, and as a result the growth of C. botulinum is inhibited (Riemann et al., 1972; Tanaka et al., 1980).

Lastly, physical controls represent another alternative to insure the botulinal safety of meat products. As reviewed by Sebranek (1979) and Sofos and Busta (1980), dehydration, freezing or refrigeration, irradiation and thermal processing have been shown to control botulinal outgrowth in laboratory studies or in certain meat products. However, the application of these factors could not be expanded to the point of absolute or sole control procedures due to a series of general/specific constraints, including: increased energy consumption; major changes in processing procedures; new legislation required; and alterations in product stability and identity (Sofos and Busta, 1980).

## D. SORBATE

In response to the recommendation by the Expert Panel on Nitrites, Nitrates and Nitrosamines that alternate preservatives to replace nitrite be evaluated (Anon., 1978a), the Monsanto Company, on April 27, 1978, filed a petition with the USDA to allow the addition of sorbic acid or

its potassium salt to bacon in conjunction with 40 mg/kg of nitrite (Anon., 1978b). The proposal stated that it allowed a reduction of nitrite in bacon along with a reduced potential for N-nitrosamine formation in frying, that the bacon produced in this manner was essentially of the same color and flavor as bacon presently available, that based on experimental data the antibotulinal protection in such bacon was at least equivalent to present commercial products and that mold inhibition during aerobic storage was improved (Anon., 1978c). On May 15, 1978 the USDA, after considering the above petition, proposed that bacon in the future be produced with 40 mg of nitrite/kg of product and 0.26% (wt/wt) potassium sorbate (Anon., 1978b). This proposal was to become effective within one year unless data submitted revealed inadequate botulism protection or N-nitrosamine formation at levels detectable by presently available techniques. However, the USDA delayed this action in order to review all research data (Berry and Blumer, 1981).

An excellent review by Sofos et al. (1979a) summarized the literature prior to 1979 which pertained to the history, chemistry, safety and preservative action of sorbate. The authors commented that the selectivity of bacterial inhibition by sorbic acid and the early reports (Emard and Vaughn, 1952; York and Vaughn, 1954; Hansen and Appleman, 1955) implicating it in enhancing or at least not restricting the growth of clostridia might have been the reasons that the compound was neither tested nor used as an antimicrobial agent in meat products until recently.

The antibotulinal efficacy of sorbic acid and/or its potassium salt, when incorporated into cure formulations alone, has been demonstrated

in several meat products. Tompkin et al. (1974) were one of the first to report the ability of potassium sorbate alone to inhibit C. botulinum growth and toxinogenesis. They evaluated cooked, uncured sausage which was manufactured with and without 0.1% potassium sorbate, inoculated with 32-38 spores/g product and then temperature abused at 27°C. Toxin was detected within 4 days in the product without sorbate, but not until 10 days in samples with sorbate (Tompkin et al., 1974). The significant ($p < 0.01$) effect of sorbic acid/potassium sorbate in retarding gas production, i.e., package swelling, and toxin production has been demonstrated in bacon (Ivey et al., 1978; Sofos et al., 1980b), chicken frankfurters (Robach et al., 1978a; Gray et al., 1979; Sofos et al., 1979b,c; Huhtanen and Feinberg, 1980), and turkey frankfurters (Huhtanen and Feinberg, 1980).

Levels of sorbate incorporated into such products range from 0.1 to 0.39%, and most authors noted an increased inhibition with increased sorbate concentration. Ivey and Robach (1978), in a study of canned, comminuted cured pork reported that sorbate concentration was significantly related to inhibition of C. botulinum, but through the fourth power of the sorbate level. In general, 0.2% sorbic acid or 0.26% potassium sorbate incorporated into a product alone is believed to be as effective as 156 mg/kg nitrite in delaying C. botulinum growth and toxinogenesis (Ivey and Robach, 1978; Gray et al., 1979; Sofos et al., 1979c).

Ivey et al. (1978) observed no significant ($p < 0.01$) difference in inhibition between sorbate-treated bacon which had no nitrite and that which had 40 mg/kg nitrite. They concluded that in bacon, inhibition of C. botulinum was due to sorbate alone and not due to a synergistic

combination of nitrite and sorbate. This contrasts with other studies that support such a synergistic interaction. In their study of chicken frankfurter-type emulsions, Sofos et al. (1979c) reported that low nitrite concentrations (20 and 40 mg/kg) did not influence C. botulinum growth and toxin production, but the addition of sorbic acid (0.2%) to these nitrite levels resulted in a significant extention of the time necessary for toxin to develop. They also observed that nitrite concentrations of 156 mg/kg or sorbic acid at 0.2% level doubled the time necessary for botulinal toxin production. The magnitude of toxin production delay was increased five-fold when 156 mg/kg nitrite and 0.2% sorbic acid were combined (Sofos et al., 1979c). Similar findings have been reported in chicken frankfurters (Gray et al., 1979; Sofos et al., 1979b), canned, comminuted cured pork (Ivey and Robach, 1978), and bacon (Sofos et al., 1980b). Furthermore, it has been demonstrated that use of 0.2% sorbic acid or 0.26% potassium sorbate along with reduced nitrite levels (20 mg/kg) would be as effective, if not more effective, than 156 mg/kg nitrite in the inhibition of C. botulinum (Ivey and Robach, 1978; Robach et al., 1978a; Sofos et al., 1979c). Evidently, cured meats which are botulinal safe can be produced using sorbate and low or even zero levels of nitrite.

Sofos et al. (1979b) discussed the possible mechanisms for the inhibitory effects of nitrite and sorbate. They postulated that the synergistic interaction between nitrite and sorbate was due to the sum of their individual effects. Sorbic acid decreases spore germination and retards cell development, while nitrite inhibits outgrowth of the germinated spores. In addition, sorbate delays the depletion of the

residual nitrite, which may increase the inhibitory effects of nitrite itself and explains why combination (nitrite-sorbate) treatments have greater stability (Sofos et al., 1979b,c; 1980a).

The hydrogen ion concentration (pH) in a product is a major influence on the antibotulinal efficacy of sorbate because the undissociated acid form is responsible for the inhibition of C. botulinum (Huhtanen and Feinberg, 1980). Using nitrite-free poultry frankfurters, Huhtanen and Feinberg (1980) demonstrated that acidification to pH 5.4-5.7 (using $H_3PO_4$ or glucono-delta-lactone) combined with 0.2% or 0.4% sorbic acid resulted in increased mean swell times. Sofos et al. (1980a) reported that the inhibition of C. botulinum spore germination, outgrowth and toxin production by sorbic acid (0.2%) in mechanically deboned chicken meat (MDCM) frankfurter-type emulsions was pH dependent. The inhibitory effect of nitrite was not influenced by pH (5.93-6.93), but when sorbate was incorporated into a product alone, it was not effective against C. botulinum at pH values above 6.0. The inclusion of nitrite in the formulation increased the effective pH (6.2) for sorbic acid inhibition of toxin production (Sofos et al., 1980a). Other factors which could influence and/or alter the antibotulinal efficacy of sorbate include: (a) use of germinated inoculum which sorbate is less effective against, (b) use of high inoculum levels, (c) variations in the precise level of cure ingredients applied to the product, or (d) growth of indigenous microorganisms (Sofos et al., 1980b).

Besides its effect on C. botulinum, sorbate has been reported to inhibit mold and yeast growth in butter (Kaul et al., 1981) and in cheese (Melnick et al., 1954a,b; Smith and Rollin, 1954). It also

reduces total microbial growth in bacon (Ivey et al., 1978a; Sofos et al., 1980b), chicken frankfurter emulsions (Sofos et al., 1979b, 1980a) and cooked, uncured sausage links (Tompkin et al., 1974). At present, the only approved application of sorbic acid in meat products is that of dipping the dry-sausage casings in a 2.5% potassium sorbate solution to inhibit mold growth on the surface of the product during the drying or aging period (Sofos et al., 1979a). In a similar application, Baldock et al. (1979) demonstrated that spraying country-cured (aged) hams with a 10% potassium sorbate solution effectively inhibited fungal development over a 30 day storage period. They observed that 400-500 mg/kg was the minimum sorbate concentration on the surface of the ham which limited mold growth, and that an initial level of 2400 mg/kg was needed to assure adequate residual concentration after 30 days. Similar results were reported by Kemp et al. (1979) with boneless, dry-cured hams dipped in a 2.5% sorbate solution.

Robach and Ivey (1978) and Cunningham (1979) evaluated the antimicrobial efficacy of potassium sorbate dips on freshly processed poultry parts. A 10% potassium sorbate solution resulted in an organoleptically acceptable product which exhibited lower total plate counts, slower rate of growth for the salmonella and reduced development of microbially-induced off-odors (Robach and Ivey, 1978; Cunningham, 1979). When whole turkey breasts and sliced turkey breast luncheon meat were processed with 0.12% sorbic acid, the psychrotrophic plate counts were extensively reduced and shelflife (at 4°C) extended from two weeks to over six weeks (Robach et al., 1980b).

The effect of sorbate on specific organisms has also been well-documented. Using a tryptic soy broth, LaRocco and Martin (1981) demonstrated the effectiveness of 0.3% sorbate combined with 3% NaCl in the inhibition of Salmonella tryphimurium growth. When bacon was prepared with 0.13 or 0.26% sorbate alone, Staphylococcus aureus growth was delayed for 14 days at 27°C storage (Pierson et al., 1979). However, these authors observed that when stored at 13°C, bacon containing both nitrite and sorbate exhibited lower numbers of staphylococci after 7 days than did bacon containing potassium sorbate alone. Tompkin et al. (1974) reported that the incorporation of 0.1% sorbate alone in cooked, uncured sausage links markedly retarded the growth of salmonella, S. aureus and Clostridium perfringens.

The combination of sorbate with reduced levels of nitrite not only provides a botulinal safe product, but also reduces the incidence of N-nitrosamine formation. Robach et al. (1980c) prepared pork bellies under commercial conditions using various nitrite and sorbate treatments and then analyzed the fried samples for volatile N-nitrosamines. They reported that bacon processed with 40 mg/kg nitrite and 0.26% potassium sorbate contained an average of 8.7 µg/kg NPYR at zero time (after slicing) and 5.4 µg/kg after 21 days at 3°C. The bacon made with 120 mg/kg nitrite contained an average of 28.2 µg/kg NPYR at zero time and 16.2 µg/kg after 21 days at 3°C (Robach et al., 1980c). Similar results were reported by Ivey et al. (1978). Tanaka et al. (1978) studied the mechanism behind the inhibition of N-nitrosamines formation by sorbate. They reported that sorbate is similar to ascorbate in that it binds nitrite (producing an oxime), and thereby makes nitrite unavailable for

N-nitrosation reaction. Sorbic acid inhibited the "in-vitro" formation of NDMA to the same extent as ascorbate, but sorbate exhibited little to no inhibitory action against the N-nitrosation of morpholine and N-methyl-aniline (Tanaka et al., 1978).

Sensory evaluation of sorbate-treated products has been favorable. Price and Stevenson (1979) studied the effects of potassium sorbate and sodium nitrite on bacon color and flavor during storage at 5 ± 2°C. They found no significant differences ($p < 0.05$) in color or taste panel scores of bacon manufactured with a nitrite input of 120 mg/kg versus bacon manufactured with a nitrite input of 40 mg/kg in conjunction with 0.26% potassium sorbate. Similarly, Paquette et al. (1980) reported that bacon formulated with 0.26% potassium sorbate in combination with 40 or 80 mg/kg sodium nitrite was not significantly different ($p < 0.05$) from samples formulated with 120 mg/kg of sodium nitrite and no potassium sorbate for color and sensory qualities. Ivey et al. (1978) evaluated bacon prepared with 0 and 40 mg/kg nitrite with and without 0.13 and 0.26% potassium sorbate. In their report, the flavor panel evaluations indicated that potassium sorbate decreased preference slightly, using experienced judges. However, consumer acceptability of bacon would probably not be affected by addition of sorbate since the panel did not consistently judge the sorbate treatments as less desirable (Ivey et al., 1978).

Despite the many beneficial attributes of sorbate, it has problems and disadvantages also. Sensory panelists who evaluated bacon processed with 40 mg/kg nitrite and 0.26% potassium sorbate have detected "chemical-like" flavors and "sweet aromatic" aromas (USDA, 1970; Berry and Blumer, 1981). In both studies, these comments were accompanied by complaints

of prickly mouth sensations, skin and throat irritations, facial swelling, numbness, and reddening of the hands. Concern that the physiological symptoms listed above were toxic/allergic responses to sorbate was alleviated by a report by Robach and Adams (1979). They reviewed the USDA study and concluded that two factors may have cuased the reported reactions: (1) the amount of bacon the panelists were exposed to was extremely large (up to 30 slices per day), and (2) the fact that one member of the USDA panel openly discussed a reaction (throat irritation) with the other members led to a psychological preconditioning of the other panelists.

Sorbate itself does not appear to be harmful when incorporated into meat products at levels suggested by meat research. Deuel et al. (1954) reported that sorbic acid was harmless to rats and dogs when incorporated in the diets to the extent of 5%, and that sorbic acid was far less toxic than sodium benzoate (a fungistatic agent similar to sorbate). Recent research (Gaunt et al., 1975) has not detected any carcinogenic effects on the part of sorbic acid even when fed at levels of up to 10% of the diet, i.e., an approximate intake of 5 g/kg/day. No effects were found at a dietary level of 1.5%, establishing this as a no-effect level equivalent to an intake of 750 mg/kg/day (Gaunt et al., 1975).

Despite the apparent safety of sorbate alone, there is some concern about the formation of mutagenic/carcinogenic compounds from the reaction between sorbic acid and sodium nitrite added to meat products. Using model systems, Namiki and Kada (1975) and Hayatsu et al. (1975) reported that excessive concentrations of sorbic acid and sodium nitrite (130,000 mg/kg) will react under highly acid conditions (pH 1.0) to form

ethylnitrolic acid (ENA). This product caused damage to bacterial DNA, i.e., mutagenic effect, in the "recombination assay" which used wild and recombinationless strains of Bacillus subtilus (Namiki and Kada, 1975). Subsequent research revealed that both ENA and another mutagenic substance, 1,4-dinitro-2-methylpyrrole, were produced by reaction between nitrite and sorbate at pH 3.5-4.2 (Namiki et al., 1981). However, Robach et al. (1980a) reported that under acidic conditions (pH 3.4) ENA is not formed at nitrite levels below 250 mg/kg and is not formed at all at meat pH (6.0) even when higher levels of nitrite (500 mg/kg) are present. ENA could not be formed in cured meat or curing brine by any mechanism so far explored; nor could ENA survive cooking temperatures, since it decomposes to acetic acid and nitrogen oxides in less than one second at 170°C (DiFate, 1978). Similarly, mutagenic 1,4-dinitro-2-methylpyrrole would not form in a meat-curing situation because low pH (3.5) and excess nitrite are necessary for the pyrrole to form, and the presence of ascorbate prevented expression of mutagenic activity (Robach et al., 1980a; Namiki et al., 1981). Therefore, it was concluded that the levels of sorbate and nitrite proposed for use in cured meats (0.26% sorbate and 40 mg/kg nitrite), together with a pH of at least 5.5 and the presence of ascorbate do not pose a hazard in regard to the formation of reaction products in cured meat or the curing brine (DiFate, 1978; Robach et al., 1980a).

Compared to other alternatives, the usage of nitrite-sorbate mixtures to protect cured meats against botulinal toxicity is an attractive alternative due to the following factors (Anon., 1976): (a) with lower nitrite levels (e.g., 40 mg/kg) the nitrosamine formation potential

would be minimized; (b) C. botulinum would be inhibited at least as well or even better compared to present formulations; (c) the low nitrite used would still give the characteristic cured meat color and flavor; (d) the shelflife of the products would increase; (e) sorbate is a naturally-occurring material which would not cause health problems, as it is metabolized like any other fatty acid in foods; (f) it is already on the GRAS list, and (g) the current processing procedures would not have to be changed.

# III. TURKEY BOLOGNA STUDY

## A. EXPERIMENTAL

The turkey bologna study was divided into two sections. The initial research, henceforth referred to as Test 1, determined the minimum level of sodium nitrite which provided an acceptable turkey bologna product according to organoleptic evaluation and chemical analyses. The second half of the study (Test 2) evaluated turkey bologna prepared with potassium sorbate, alone or in combination with reduced levels of sodium nitrite, according to organoleptic, chemical and microbiological methods.

### 1. Bologna Preparation

Identical techniques were used in the preparation of turkey bologna for both Tests 1 and 2. Deboned turkey thigh pieces and turkey adipose tissue were purchased as needed from a commercial processor (Bill-Mar Foods). Random samples of lean and adipose were analyzed for percent moisture, fat and protein by standard procedures (AOAC, 1975). Formulations based on the analyses of the raw materials were computed to yield bologna with 25% fat and 10% added moisture in the finished product based on a 90% anticipated smokehouse yield.

Appropriate amounts of adipose tissue (turkey fat), turkey meat and ice water were chopped to a uniform paste in a vertical cutter mixer (Hobart, Model VCM 40E) with a 40 liter nominal capacity. Formulated amounts of spices, including sugar and salt (4.5% of the finished

product (wt/wt), sodium ascorbate (250 mg/kg) and curing phosphates (0.25%, an equimixture of sodium tripolyphosphate, sodium pyrophosphate, sodium hexametaphosphate and monosodium phosphate; Griffith Labs., Chicago, IL.) were added to the meat-fat system and mixed for 80 seconds at high speed. The batter was transferred to a 80 kg capacity Mincemaster for emulsification. Batches (7.0 ± 0.5 kg each) of the batter were then placed in the vertical cutter mixer and the prescribed quantities of sodium nitrite (0 to 156 mg/kg) and/or potassium sorbate (0 to 0.39% wt/wt) were added (Tables 2 and 3). The mixture was chopped at low speed for 40 seconds under vacuum to obtain uniform blending of the additives.

Table 2: Target nitrite levels added to turkey bologna for Test 1.

| Treatment No. | Sodium nitrite (mg/kg) |
|---|---|
| 00 | 0 |
| 20 | 20 |
| 40 | 40 |
| 60 | 60 |
| 100 | 100 |
| 156 | 156 |

Table 3: Target nitrite-sorbate concentrations added to turkey bologna for Test 2.

| Treatment No. | Sodium nitrite (mg/kg) | Potassium sorbate (%) |
|---|---|---|
| 00 | 0 | 0.0 |
| 02 | 0 | 0.26 |
| 03 | 0 | 0.39 |
| 40 | 40 | 0.0 |
| 60 | 60 | 0.0 |
| 42 | 40 | 0.26 |
| 62 | 60 | 0.26 |
| 156 | 156 | 0.0 |

The temperature of the blends was recorded (range for Test 1, 11-16°C and for Test 2, 4-6°C). The batter was transferred to a water pressure sausage stuffer (E. F. Zuber, Minneapolis, MN.), stuffed into 80 mm bologna casings (cellulose) and cooked in a smokehouse equipped with temperature and humidity controls (Drying Systems, Inc., Chicago, IL.). No smoke was added during the heat process listed in Table 4.

Table 4: Cooking schedule for turkey bologna (Tests 1 and 2).

| Time (hours) | Dry Bulb (°C) | Wet Bulb (°C) | R.H. (%) |
|---|---|---|---|
| 2 | 60 | 36 | 30 |
| 2 | 71 | 47 | 30 |
| *1 | 77 | 70 | 80 (steam) |

*One hour or time necessary to reach 68.8°C internal temperature.

The product was cooked to a minimum internal temperature of 68.8°C (156°F) followed by a cold water shower cooking for 20 minutes. The samples were allowed to cool at room temperature for 2 hours and then held overnight at 4°C. Weights of the individual bologna chubs were recorded before and after cooking with resultant yields of 80 to 89% for Test 1 and from 91 to 94% for Test 2. The casings were removed and 1 kg sections of the bologna were vacuum packaged (Multivac, West Germany) in Van 4® bags (Koch, Kansas City, MO.) for three weeks of storage in the dark at 4 ± 1°C.

2. Organoleptic Analysis

In the initial study (Test 1), replicate panels consisting of 20 untrained judges evaluated the meat samples at regular intervals over

the three week storage period. Samples were evaluated under white light for color, odor, flavor and overall acceptability using the method outlined by Stone et al. (1974). An unstructured interval scale of 170 mm was used for each attribute. Anchor points were placed 20 mm from each end of the line and were labelled with the weakest attribute on the left and the strongest on the right. A sample of the evaluation form is found in the Appendix. Within each treatment group, bologna slices (7 mm thick) were randomly selected and cut into 3 mm squares. The unheated meat samples were placed in randomly numbered plastic petri dishes and presented to the panelists. The judges were asked to compare each sample to the reference sample (R) which was prepared with 156 mg/kg sodium nitrite. Panel scores were measured from the left (0 mm) on each line on the evluation form and were recorded in mm for subsequent statistical analysis.

For Test 2, an unreplicated panel of 10 trained judges examined the bologna samples over the three week storage period. These 19 panelists were selected as the most consistent and/or perceptive of the 20 judges in Test 2. All other sensory procedures were the same as those described above.

3. Analytical Methods

Proximate analysis for percent moisture, fat and protein in the raw turkey thigh meat and the 0 mg/kg sodium nitrite turkey bologna were determined by the AOAC official methods (1975).

The residual nitrite level of all bologna samples was ascertained at regular intervals over the three week storage period using the AOAC

official method (1975). Since α-naphthylamine was utilized in this analysis, caution was exercised during the process due to the carcinogenicity of the compound. Two replicates per treatment group were analyzed.

Thiobarbituric acid (TBA) values were determined on ground bologna samples at regular intervals during the three weeks of storage using the method of Tarladgis et al. (1960), as modified by Zipser and Watts (1962). Analyses included four distillations per sample and two colormetric reactions per distillation. Absorbance was read at 532 nm and TBA numbers (mg malonaldehyde per 1000 g of sample) calculated using a constant of 7.8.

Bologna color was objectively evaluated at regular intervals over the three week storage period using a Hunter D-25 Color Difference Meter (Hunter Associates Lab., Fairfax, VG.). The standard pink reference tile had the following values: L=67.6, $a_L$=21.4 and $b_L$=11.9. A randomly selected slice (8 mm in diameter, 7 mm thick) from each treatment group was placed (flat) in an optically inert glass dish, and readings taken before and after turning the dish 90°. The slice was inverted and the process repeated to give a total of four replications per sample.

pH was determined for ground turkey bologna homogenates (25 g meat in 25 ml deionized water) using a Corning 10 pH meter. Two replicates per treatment group were analyzed.

4. Statistical Analysis

Sensory evaluation data was analyzed as a factorial design according to Gill (1978). Three-way analysis of variance for evaluators,

treatment and time effects was performed on the panel scores for each attribute. The effect of the evaluators was included to compensate for the lack of replication in the original sample preparation. The level of significance adopted in this study was $p < 0.01$. When the treatment effect was significant, mean panel scores for each treatment group were compared with the control (156 mg/kg sodium nitrite) using Dunnett's test (Dunnett, 1955, 1964). Simple linear regression and simple linear correlation analyses were also performed between mean panel scores for various attributes (Gill, 1978), using a level of significance of $p < 0.001$.

Two-way analysis of variance for treatment and time effects was conducted on TBA values and the Hunter L, $a_L$ and $b_L$ readings. For significant ($p < 0.01$) main effects and/or interactions, the mean values from each treatment group were compared with the control (156 mg/kg sodium nitrite) using Dunnett's test (Dunnett, 1955, 1964). Simple linear regression and simple linear correlation analyses were also conducted between sensory evaluation data, TBA values and Hunter L, $a_L$ and $b_L$ readings.

5. N-Nitrosamine Analysis

The possible presence of volatile N-nitrosamines in turkey bologna samples from Tests 1 and 2 was determined using the gas chromatograph-Thermal Energy Analyzer (TEA) method of Fine et al. (1975), as modified by Robach et al. (1980c). Twenty-five grams of cold, ground bologna were placed in a distillation flask, mineral oil was added and distillation commenced. After extraction of the distillate with dichloromethane, the

solution was poured into a "Preptube" (Thermo Electron Corporation, Waltham, MS.) which had been previously washed with 50-60 ml of dichloromethane, and collected in a Kuderna-Danish concentrating apparatus fitted with a 4 ml receiver. The sample was concentrated and brought to 0.5 ml with dichloromethane, transferred to a 1 ml conical-shaped vial with a Teflon-lined cap and placed in a freezer (-30°C).

Quantitative determination of the volatile N-nitrosamines was carried out using a GC-TEA system comprised of a Hewlett-Packard Model 5710-A gas chromatograph coupled to a TEA Model 502/LC (Thermo Electron Corporation, Waltham, MS.) via a 1/8 inch glass-lined stainless steel transfer line. The GC column was a 4 m X 3 mm i.d. glass column packed with 10% Carbowax 20M and 5% KOH on 80/100 mesh Chromosorb W (Varian Associates). Operating conditions for this system were: GC carrier gas and flow rate, helium at 40 ml/min; GC injection port temperature, 150°C; GC column temperature, 180°C, isothermal; TEA pyrolyzer furnace, 425°C; TEA reaction chamber pressure, 1.5 Torr; TEA attentuation, as appropriate; ice bath temperature, -160°C (isopentane/liquid nitrogen slush bath); GC-TEA heated transfer line, 175°C. Raw data were collected and processed by a Hewlett-Packard Model 3353 Lab Automation System. A Linear Instruments Model 361 strip chart recorder was used to record the chromatograms.

A stock standard N-nitrosamine solution was obtained from the Illinois Institute of Technology, Research Institute, Chicago. The solution was a six component mixture containing nominally 50 mg/ml of each of N-nitrosodimethylamine (NDMA), N-nitrosodiethylamine (NDEA), N-nitrosodibutylamine (NDBA), N-nitrosopiperidine (NPIP), N-nitrosopyrrolidine

(NPYR) and N-nitrosomorpholine (NMOR). The standard was stored in a freezer maintained at about -30°C. The limits of detection (defined as 10 X baseline noise) for the various N-nitrosamines were: NDMA, 0.2 µg/kg; NDEA, 0.2 µg/kg; NDBA, 0.5 µg/kg; NPIP, 0.5 µg/kg; NPYR, 0.5 µg/kg; and NMOR, 0.5 µg/kg.

Concentration of the N-nitrosamine of interest was calculated from the following equation:

$$\mu g/kg = \frac{1000 \cdot V \cdot X \cdot C}{S \cdot W}$$

where V = final volume of extract after concentration, in ml; X = sample peak height (area); C = standard concentration, mg/ml; S = standard peak height (area); and W = weight of meat sample analyzed, in grams. This equation assumes equal aliquots of standard and sample injected.

Recovery of the N-nitrosamines from the bologna was determined by spiking known amounts of each nitrosamine of interest into a distillation flask containing 25 g of bologna sample, prior to distillation. Results were compared to nonspiked samples. Recoveries of all N-nitrosamines averaged 107% for 1 µg/kg spiking, 99% for 5 µg/kg spiking and 96% for 10 µg/kg spiking. Since only the reference sample (prepared with 156 mg/kg sodium nitrite) yielded any apparent N-nitrosamines, recovery determinations were completed using 30 replicates from this treatment group.

6. Clostridium botulinum Study

The various nitrite-sorbate combinations in the turkey bologna of Test 2 were examined as to their efficacy in inhibiting C. botulinum

growth and toxinogenesis. A C. botulinum spore suspension containing equal numbers of four Type A strains (36A, 52A, 77A and 10755A) and five Type B strains (41B, 53B, 213B, 7949B and Lamanna B) was prepared as described by Rhodes and Jarvis (1976). Spore suspensions of the individual strains were supplied by the Monsanto Company (St. Louis, MO.). C. botulinum spore counts were determined using the three-tube Most Probable Number (MPN) technique, employing incubation in a TPSY broth (5% trypticase, 0.5% peptone, 1.0% yeast extraction, 0.2% sucrose and 0.1% sodium thioglycollate at pH 7.2 for 7 days at 35°C) (Emodi and Lechovich, 1969). The composite spore suspension was diluted in sterile, distillate water, and heat-shocked at 80°C for 15 minutes.

Sufficient inoculum was spread between two-50 g bologna slices to provide a concentratin of 100 spores/g of product. Inoculated and uninoculated samples from each treatment group were vacuum packaged (Multivac, West Germany) in Vac 4 ® bags (Koch, Kansas City, MO.) and temperature abused at 27 ± 1°C. Samples were removed after 0, 3, 5, 7, 10, 14, 21 and 28 days when held under abuse conditions. Three unswollen packages per treatment group were analyzed at each sampling time as long as unswollen packages remained available. If packages appeared swollen, they were removed for toxin analysis.

Assays for botulinal toxin were conducted using the procedure of Christiansen et al. (1973). Turkey bologna sample (50 g) was blended with 100 ml of gelatin--phosphate diluent (0.2% gelatin and 0.4% $Na_2HPO_4$ at pH 6.2). The homogenate was centrifuged and 0.2 ml aliquots of the supernatant was injected intraperitoneally into each of two Swiss Webster white mice weighing approximately 20 g. Death of at

least one mouse after the appearance of typical symptoms of botulism during the next 72 hours was considered evidence of the presence of botulinal toxin. Extracts from uninoculated samples stored at 27 ± 1°C were periodically injected into mice as controls

B. RESULTS AND DISCUSSION

1. Test 1: Determination of Minimum Nitrite Level

a. Organoleptic analysis: Turkey bologna chubs manufactured with 0, 20, 40, 60, 100 and 156 mg/kg of sodium nitrite were evaluated at regular intervals by sensory panels for color, odor, flavor, off-odor, off-flavor and overall acceptability. The influence of various nitrite levels on the mean panel scores for color is shown in Figure 2. Analysis of variance indicated that only sample treatment (B) was significant ($p < 0.01$) for this attribute (Appendix Table A-1). A comparison of treatment means over time (Appendix Table A-2) revealed that only the nonnitrite treated sample was significantly different ($p < 0.01$) from the control (156 mg/kg nitrite). The unpleasant grey-brown color observed in the turkey bologna prepared without sodium nitrite has also been reported in nonnitrite treated beef-pork frankfurters (Wasserman and Talley, 1972) and thuringer sausage (Dethmers and Rock, 1975). Theoretically, MacDougall et al. (1975) supported the findings of this study, that 20-25 mg/kg of nitrite was sufficient to insure an adequately stable color. Other researchers have claimed that 20 mg/kg of nitrite gave a cured color inferior to that of comminuted meat samples with 40 to 156 mg/kg of nitrite (Gray et al., 1979; Sofos et al., 1979c). A minimum of 40-50 mg/kg of nitrite for acceptable cured meat color has

Figure 2. Effect of various nitrite levels on the mean panel scores for turkey bologna color during storage at 4°C.

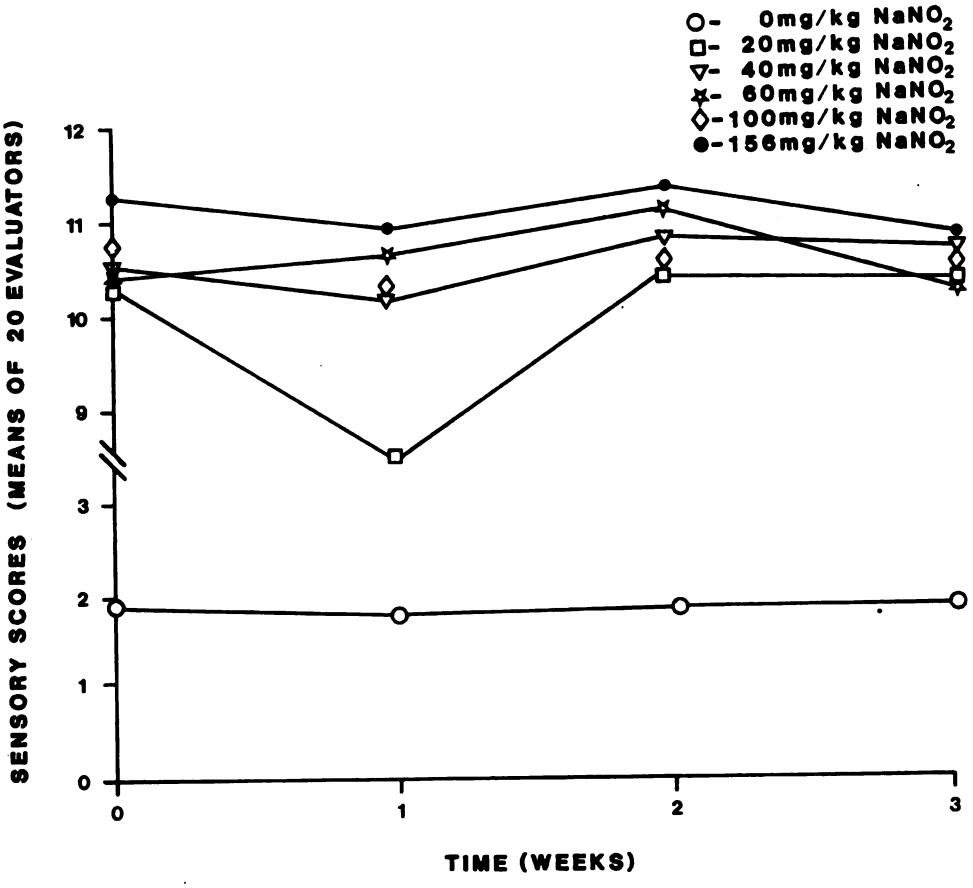

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 20 EVALUATORS)
12
11
10
9
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 2

been reported in bacon (Paquette et al., 1980) turkey frankfurters (Sales et al., 1980), beef-pork frankfurters (Sebranek et al., 1977), and corned beef briskets (Shults et al., 1977), but this color was observed to be similar to that of samples treated with 156 mg/kg of nitrite. Possible explanations for this difference in results could be: (a) various types of meat differ in their myoglobin and iron content, both of which are major determinants of color formation by nitrite, or (b) variations in product formulation techniques and/or in the homogenization of raw materials influence the distribution of nitrite. Despite the disagreement on the minimum nitrite required for color development, this study does concur with others that color intensity/panel scores for color increased with increased nitrite input in the formulation.

The influence of nitrite level on the mean panel scores for turkey bologna flavor and odor is shown in Figures 3 and 4, respectively. Analysis of variance revealed that only the sample treatment (B) was significant ($p < 0.01$) for both attributes (Appendix Tables A-3 and A-4). For the flavor scores, comparison of treatment means over time (Appendix Table A-5) indicates that samples containing 40 mg/kg of nitrite or more were not significantly different ($p < 0.01$) from the sample with 156 mg/kg of nitrite. Taste panel members commented that the nonnitrite treated samples exhibited a "turkey or poultry" flavor and that the "cured ham-like" flavor increased as nitrite input was increased. Similar observations have been reported for beef-pork frankfurters (Wasserman and Talley, 1972; Hustad et al., 1973; Simon et al., 1973; Sebranek et al., 1977), turkey frankfurters (Sales et al., 1980), chicken frankfurters (Gray et al., 1979), thuringer sausage (Dethmers

Figure 3: Effect of various nitrite levels on the mean panel scores for turkey bologna flavor during storage at 4°C.

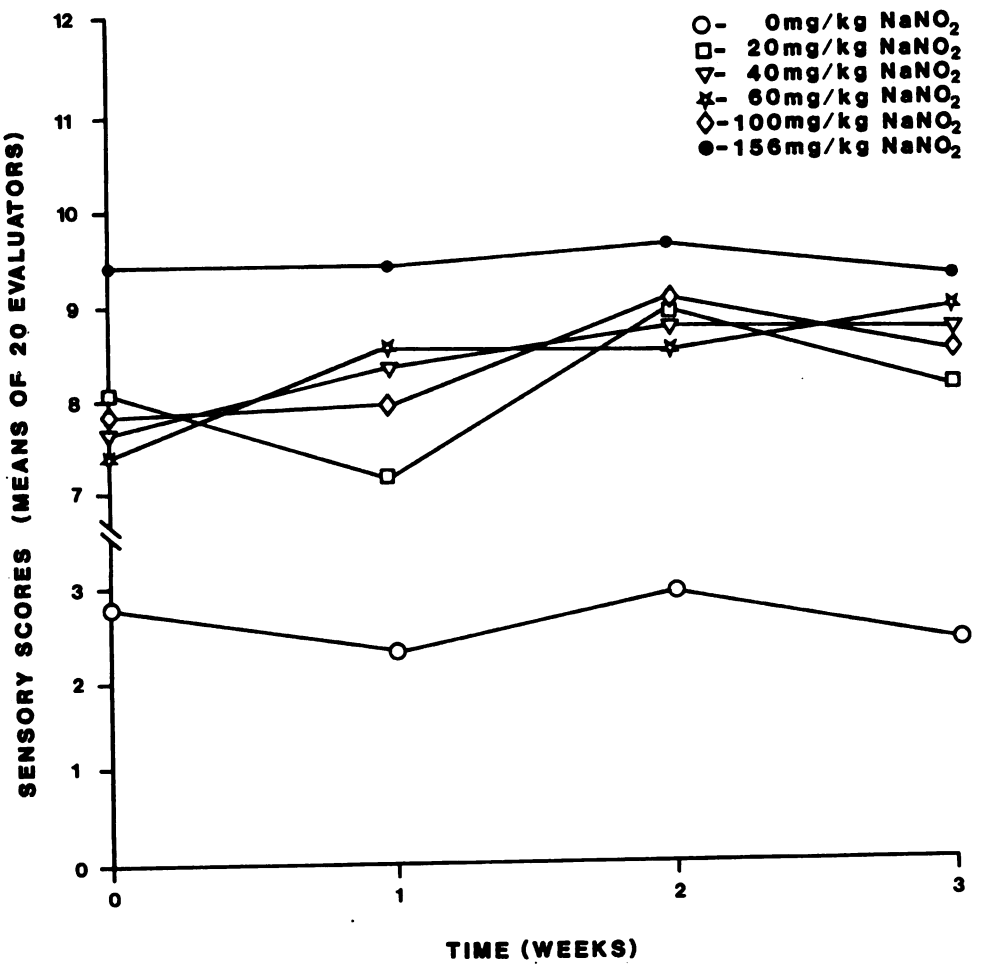

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 20 EVALUATORS)
12
11
10
9
8
7
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 3

Figure 4. Effect of various nitrite levels on the mean panel scores for turkey bologna odor during storage at 4°C.

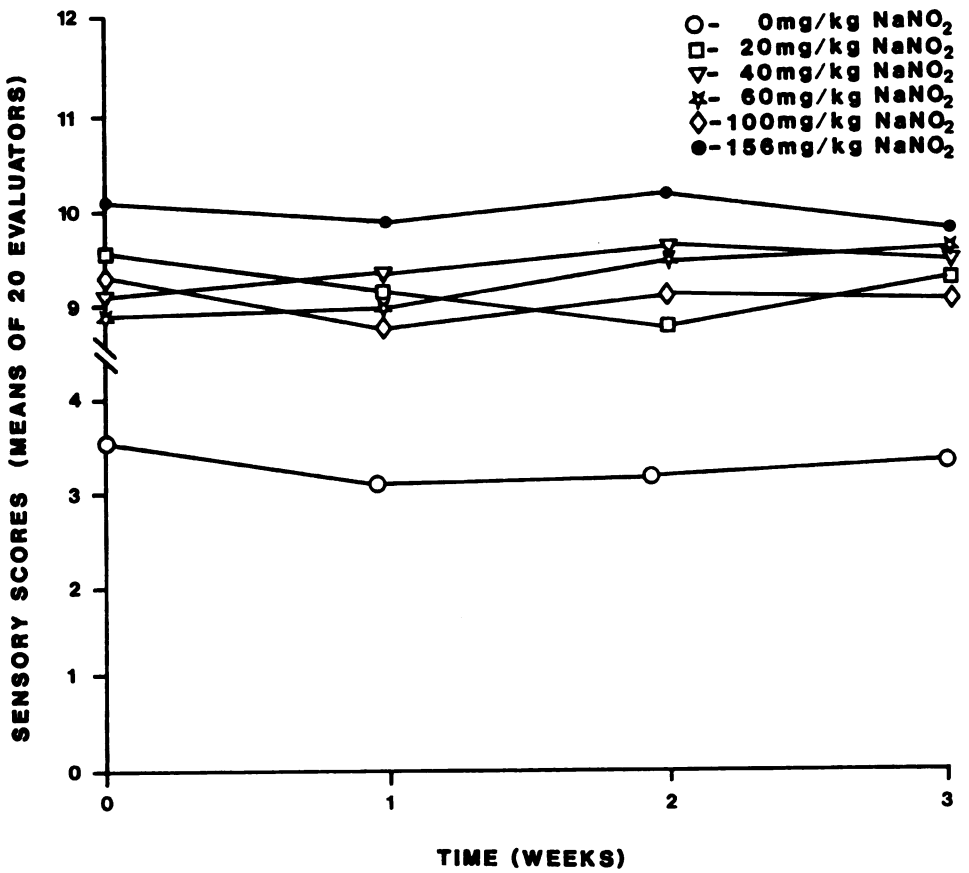

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 20 EVALUATORS)
12
11
10
9
4
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 4

and Rock, 1975) and comminuted pork (Hadden et al., 1975). In their studies on salami sausage, Skjelkvale and Tjaberg (1974) and Uram (1981) reported that panelists could not distinguish between products with and without nitrite. Therefore, other investigators have suggested that comminuted products (MacNeil and Mast, 1973; Greene and Price, 1975) and bacon (Kimoto et al., 1976) with acceptable cured flavor could be produced without the use of nitrite, providing sufficient salt and/or other flavor producing compounds were incorporated into the formulation. However, the results from the turkey bologna study emphasized that the presence of at least low levels of nitrite is a prerequisite for acceptable cured flavor development. Similarly, Wasserman and Talley (1972) noted that the base frankfurter formulation, containing a commercial spice mixture alone, was not sufficient to impart a good frankfurter flavor in the absence of sodium nitrite in the cure.

In contrast to the flavor scores, the evaluation of turkey bologna odor (Appendix Table A-6) shows that only the samples prepared without nitrite were significantly different ($p < 0.01$) from the reference (156 mg/kg nitrite). Although many studies have evaluated the contribution of nitrite to cured flavor, few have reported its effect on meat aroma. Therefore, an explanation for why the lower nitrite level (20 mg/kg) provided acceptable bologna odor, but unacceptable flavor is not readily available. One possible reason for these results is that the relatively large number of bologna samples (six) presented to the panelists overwhelmed or saturated their olfactory functions, thereby reducing the panelist's perception and/or ability to differentiate between various treatments. Secondly, many of the judges commented that the spice level

was too high in all samples. The aromatic components contributed by these spices may have masked the cured flavor in bologna prepared with 20 mg/kg of nitrite. Therefore, greater nitrite concentrations were required (>40 mg/kg) in order for the panelists to detect flavor differences between treatment groups.

An oxidized or rancid flavor is considered a desirable characteristic in country-styled ham, but is an undesirable quality if present in other cured meat products (MacDonald, 1978). Bologna samples in this study were evaluated for both off-flavor and off-odor formation during anaerobic (vacuum-packaged) storage for 21 days at 4°C (Figures 5 and 6, respectively). The terms of "off"-flavor and "off"-odor were used instead of "oxidized" or "rancid" since it is difficult to subjectively distinguish flavors resulting from lipid oxidation from those arising from other reactions such as bacterial souring. However, panelists were asked to judge the samples on the basis of off-flavor and off-odor development which closely resembled oxidized flavor. Analysis of variance indicates that only sample treatment (B) was significant ($p < 0.01$) for both attributes (Appendix Tables A-7 and A-8). Comparison of off-flavor scores (Appendix Table A-9) reveals that products prepared with at least 40 mg/kg of nitrite were not significantly different ($p < 0.01$) from the 156 mg/kg nitrite sample, while only 20 mg/kg of nitrite was necessary to significantly ($p < 0.01$) reduce detectable off-odors (Appendix Table A-10). These results were confirmed by Sales et al. (1980) who reported that rancid odor and flavor in turkey frankfurters decreased with increasing nitrite concentration and that there was no significant differences in samples with 40 and 100 mg/kg of nitrite.

Figure 5: Effect of various nitrite levels on the mean panel scores for turkey bologna off-flavor during storage at 4°C.

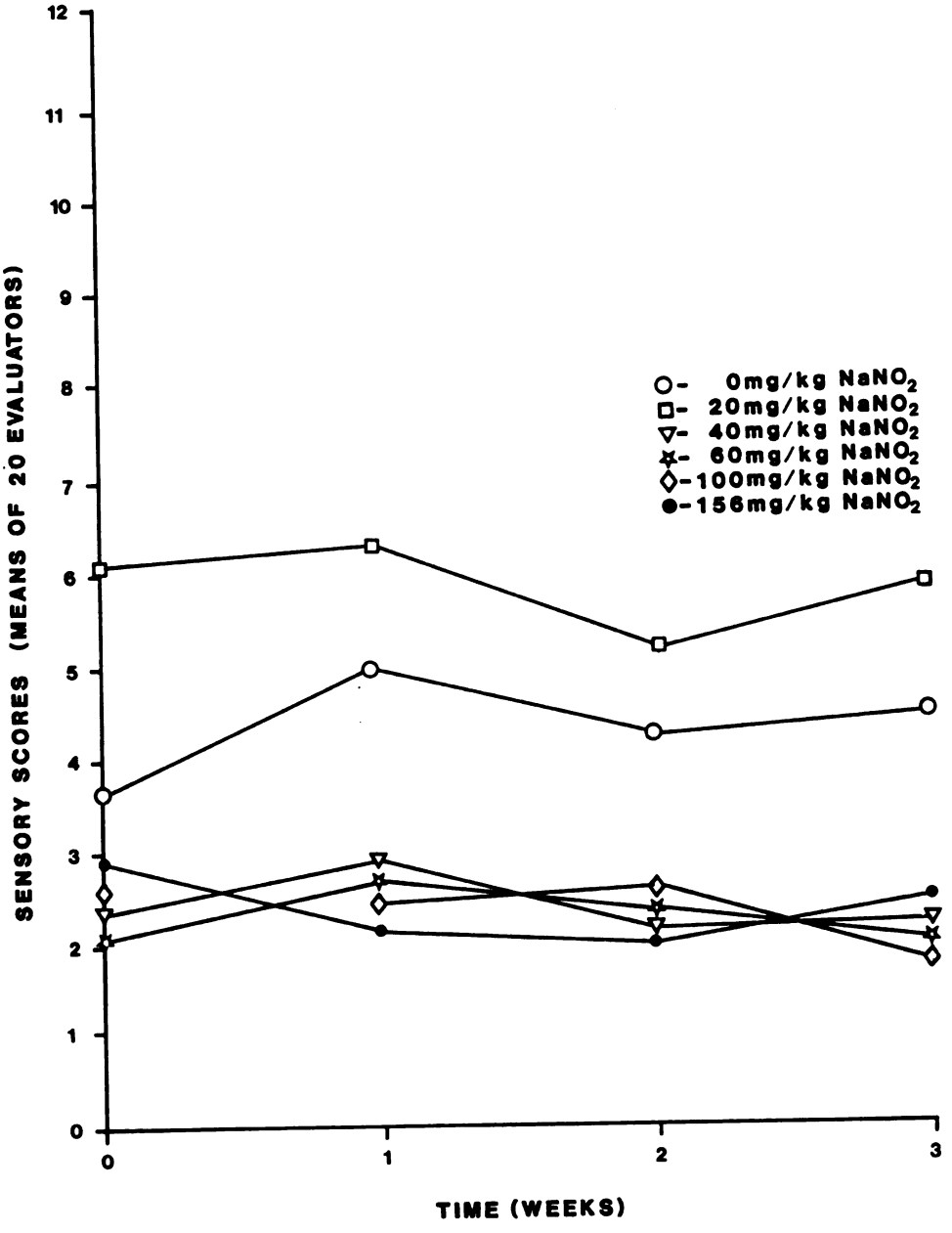

SENSORY SCORES (MEANS OF 20 EVALUATORS)
TIME (WEEKS)
0
1
2
3
0
1
2
3
4
5
6
7
8
9
10
11
12
O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2

Figure 5

Figure 6: Effect of various nitrite levels on the mean panel scores for turkey bologna off-odor during storage at 4°C.

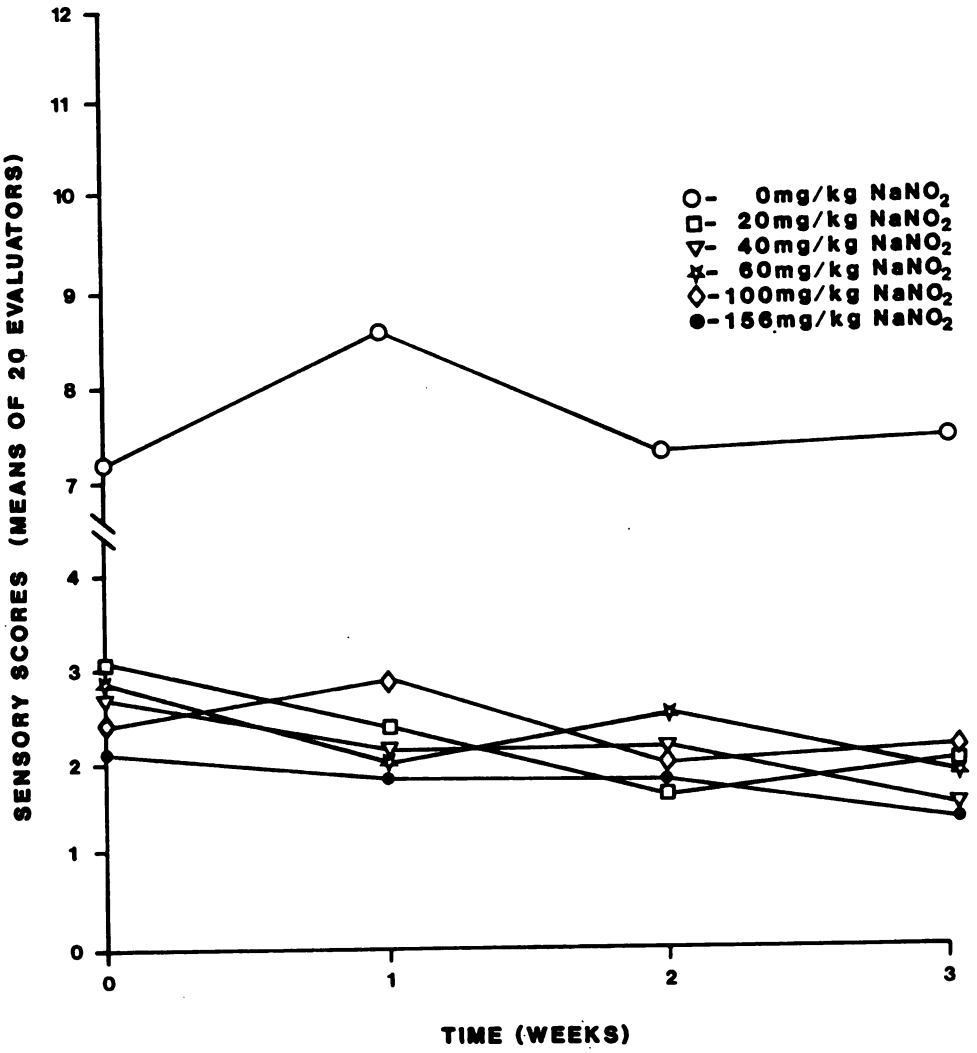

SENSORY SCORES (MEANS OF 20 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
7
8
9
10
11
12
○- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2

Figure 6

Similarly, Dethmers and Rock (1975) studied thuringer sausage and observed that the addition of nitrite in the cure reduced off-flavor development. However, these authors commented that nitrite levels beyond 50 mg/kg yielded the lowest off-flavor scores. Comparable results have been reported for various comminuted meat systems prepared with 0 and 156 mg/kg of nitrite (Hadden et al., 1973; Waldman et al., 1974; Fooladi et al., 1979; Igene et al., 1979).

The reason that 20 mg/kg of nitrite significantly reduced off-odor, but did not decrease the off-flavor detected cannot be readily answered. In explanation, nitrite may influence the volatile components which contribute to meat aroma more readily than it affects the nonvolatile compounds constituting cured meat flavor. This is substantiated by results of studies which revealed the significant ($p < 0.01$) effect exerted by nitrite on the level of carbonyls, such as hexanal, in the headspace vapor above cured and uncured comminuted pork systems (Cross and Ziegler, 1965; Hadden et al., 1973). It also could be possible that panelists are just more sensitive to changes in the relative levels of aromatic components compared to similar alterations in the flavor compounds of a meat. Another factor to be considered is that many panelists may have interpreted and/or attributed the contributions from the high spice content in bologna as off-flavor and off-odor characteristics.

The influence of nitrite level on the overall acceptability of turkey bologna is shown in Figure 7. Analysis of variance (Appendix Table A-11) indicates that only sample treatment (B) exerted a significant ($p < 0.01$) effect on this attribute. Comparison of treatment means over time (Appendix Table A-12) reveals that products prepared with

Figure 7: Effect of various nitrite levels on the mean panel scores for turkey bologna overall acceptability during storage at 4°C.

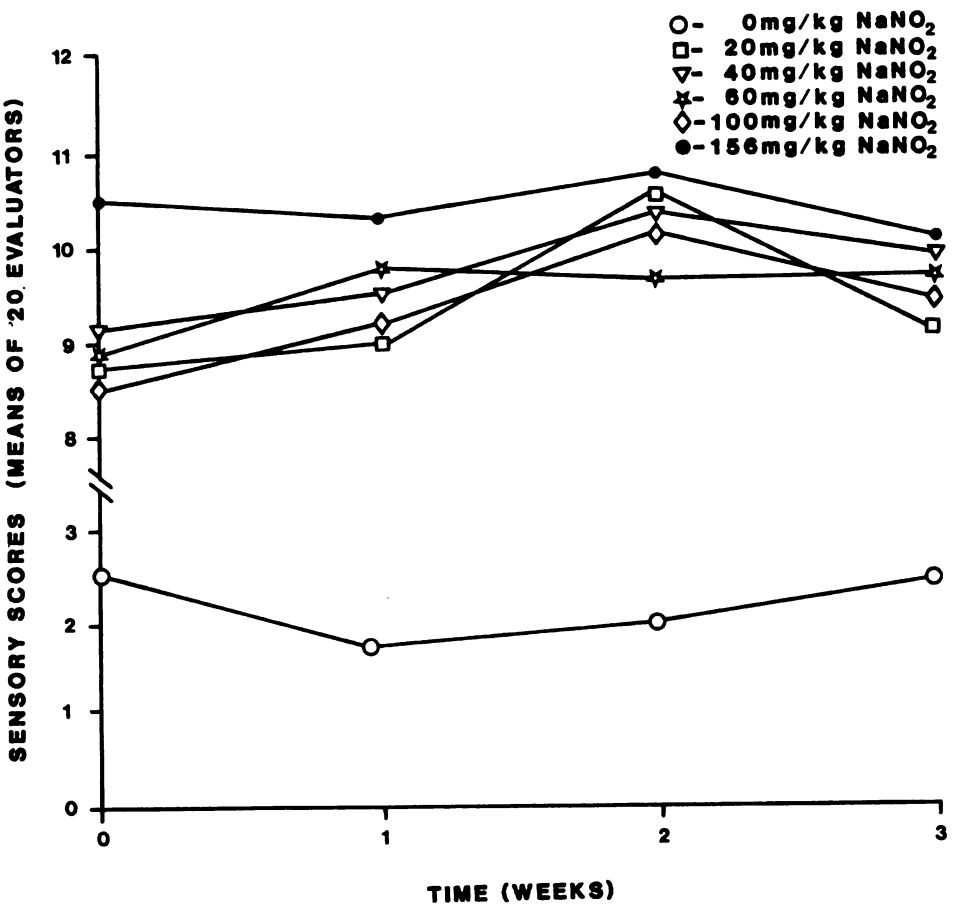

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
✲- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 20 EVALUATORS)
12
11
10
9
8
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 7

20 to 100 mg/kg of nitrite were comparable to the control (156 mg/kg nitrite). Dhillon and Maurer (1975) and Gray et al. (1979) agreed with these results, but most authors have concluded that the inclusion of nitrite in the formulation, regardless of the level of incorporation, is the determining factor in product acceptance (Sebranek et al., 1977; Paquette et al., 1980; Sales et al., 1980).

Nitrite influences many attributes of a cured meat product, but its effect on color is the most pronounced. In fact, color appeared to be the primary determinant in the acceptance of turkey bologna. In this study, red lights were not used to mask color differences between different treatment groups. Several panelists commented that they would have rated the nonnitrite treated turkey bologna more acceptable if it had not been for the unpleasant grey-brown color of the sample. Similarly, Sebranek et al. (1977) and Price and Greene (1978) have reported that sample color and appearance exert a decisive effect on a panelist's judgment. Evidently, the influence of superior ($p < 0.01$) color on a consumer panel, where color is not screened during evaluation, is sufficient to increase the overall acceptability of the product (Sebranek et al., 1977) and may also effect the scores subsequently assigned to flavor and aroma attributes of the same sample (Paquette et al., 1980).

Although several investigators have reported that panel ratings for color, aroma, flavor and acceptability declined as shelf storage increased (Simon et al., 1973; Waldman et al., 1974; Gray et al., 1979), time was not a significant factor for any of the six attributes tested in this study. Evidently, vacuum packaging the turkey bologna, using films of low oxygen permeability, reduced the oxidative and/or degradative

reactions which can result in color and/or flavor loss and off-flavor and/or off-odor development during storage.

For the most part, the presence of nitrite in the bologna samples was associated with increased color, cured flavor and aroma, and decreased off-flavor and off-odor production. However, increased nitrite input did not always result in improved sensory scores. Although this inconsistent trend was probably due to product variation and/or inexperience of panel members, it serves to emphasize that turkey bologna prepared with reduced levels of nitrite is comparable to the reference with 156 mg/kg of nitrite.

Simple linear correlation and simple linear regression analyses were performed between the mean panel scores of various sensory attributes. Table 5 indicates that increasing flavor/odor intensity was correlated with decreasing off-flavor/off-odor scores. In addition, increasing cured odor/flavor intensity or decreasing off-odor/off-flavor intensity was associated with greater product acceptability. Price and Greene (1978) reported that overall flavor desirability ratings were correlated significantly ($p < 0.02$) with rancid flavor scores ($r = 0.89$). A high, but not significant r value (0.81) was obtained between cured meat flavor scores for overall flavor desirability. From these facts it was concluded that "cured" flavor was not directly related to a lack of "oxidized" flavor, but a "desirable" flavor appeared to be associated with a more intense cured flavor and a less intense rancid flavor (Price and Greene, 1978).

It should be noted in Table 5 that correlations and regressions involving the off-flavor attribute are all less significant ($p < 0.05$)

Table 5: Linear correlation and regression of various sensory attributes of turkey bologna containing various nitrite treatments.

| Contrast | | Regression | Correlation | |
|---|---|---|---|---|
| Flavor | VS Acceptability | Y = -0.91 + 1.23 X | + 0.99 | *** |
| Color | VS Acceptability | Y = 0.59 + 0.86 X | + 0.98 | *** |
| Odor | VS Acceptability | Y = -1.86 + 1.23 X | + 0.98 | *** |
| Off-Flavor | VS Acceptability | Y = 10.78 - 0.76 X | - 0.37 | ** |
| Off-Odor | VS Acceptability | Y = 12.48 - 1.37 X | - 0.98 | *** |
| Off-Flavor | VS Off-Odor | Y = 1.25 + 0.55 X | + 0.37 | ** |
| Off-Flavor | VS Flavor | Y = 9.70 - 0.68 X | - 0.41 | ** |
| Off-Flavor | VS Odor | Y = 10.11 - 0.58 X | - 0.35 | ** |
| Off-Odor | VS Odor | Y = 11.50 - 1.08 X | - 0.97 | *** |
| Off-Odor | VS Flavor | Y = 10.84 - 1.10 X | - 0.98 | *** |
| Odor | VS Flavor | Y = -0.68 + 0.99 X | + 0.98 | *** |

***($p < 0.001$)
**($p < 0.05$)

than the rest. The mean scores for this attribute did not provide a consistent trend. Samples prepared with higher levels of nitrite were considered to exhibit more off-flavor than bologna containing low nitrite levels; and in particular the 20 mg/kg of nitrite product received greater scores for rancidity than the nonnitrite treated sample.

b. Proximate analysis and pH: The proximate analyses of raw turkey meat and of turkey bologna prepared without sodium nitrite (0 mg/kg) are listed in Table 6. These results are consistent with the moisture, fat and protein levels reported by others for turkey thigh meat (Tompkin et al., 1978d; Uebersax et al., 1978) and turkey frankfurters (Sales et al., 1980). The fat content of the turkey bologna sample was 9% below the target level of 25%, and therefore the product had greater moisture and protein levels than similar comminuted poultry products

which contained high fat concentration (Froning et al., 1971; Gray et al., 1979). It is interesting to note that Baker et al. (1969) observed that as the level of fat in a chicken frankfurter formulation is increased, tenderness and juiciness of the product decreased, but the flavor was not affected.

Table 6: Proximate analysis of raw turkey thigh meat and nonnitrite treated turkey bologna.

| Sample | Moisture (%) | Fat (%) | Protein (%) |
|---|---|---|---|
| Raw meat | 73.80 ± 0.84 | 5.86 ± 1.00 | 15.80 ± 1.70 |
| Turkey bologna | 60.90 ± 0.02 | 16.15 ± 0.37 | 15.60 ± 0.72 |

The pH readings (Table 7) for turkey bologna decreased slightly over the three week storage period regardless of the added nitrite level. The mean pH values tended to increase with increased nitrite input. All of the bologna samples exhibited higher pH values than have been reported for other comminuted meats (Hill et al., 1973; Waldman et al., 1974; Hargett et al., 1980). This was probably because an acidulant, such as glucono-delta-lactone, was not incorporated into the turkey product formulation.

c. Residual nitrite: Table 8 lists the residual nitrite analysis of turkey bologna chubs prepared with various nitrite levels. Although the level of residual nitrite increased with increased nitrite input, the 156 mg/kg sample only contained a range of 49-58 mg/kg of residual nitrite. Similar results have been reported in studies of thuringer

Table 7: pH values for turkey bologna prepared with various levels of sodium nitrite and stored at 4°C over three weeks.

| Nitrite (mg/kg) | Time (Weeks) 0 | 1 | 2 | 3 | X |
|---|---|---|---|---|---|
| 00 | 6.25 | 6.20 | 6.20 | 6.15 | 6.20 ± 0.04 |
| 20 | 6.23 | 6.23 | 6.20 | 6.15 | 6.20 ± 0.04 |
| 40 | 6.29 | 6.30 | 6.20 | 6.18 | 6.24 ± 0.06 |
| 60 | 6.25 | 6.27 | 6.25 | 6.20 | 6.24 ± 0.03 |
| 100 | 6.25 | 6.25 | 6.20 | 6.23 | 6.23 ± 0.02 |
| 156 | 6.27 | 6.30 | 6.23 | 6.20 | 6.25 ± 0.04 |

sausage (Dethmers and Rock, 1975), beef-pork bologna (Lin and Sebranek, 1979), beef-pork frankfurters (Fiddler et al., 1972; Hustad et al., 1973; Waldman et al., 1974; Hargett et al., 1980), cooked salami (Uram et al., 1981) and chicken frankfurters (Gray et al., 1979; Sofos et al., 1979c). The majority of these authors concluded that with a 40 mg/kg nitrite input, a 60-80% loss of nitrite results during processing, while a 40-50% loss occurs at the 156 mg/kg level.

Table 8: Residual nitrite analysis of turkey bologna prepared with various nitrite levels and stored at 4°C.

| Time | Nitrite Treatment (mg/kg) 00 | 20 | 40 | 60 | 100 | 156 |
|---|---|---|---|---|---|---|
| Week 0 | 10.5* | 9.5 | 22.0 | 32.0 | 48.0 | 49.0 |
| Week 1 | 8.5 | 12.0 | 20.5 | 29.0 | 46.5 | 47.0 |
| Week 2 | 6.0 | 10.3 | 21.0 | 26.8 | 42.0 | 58.0 |
| Week 3 | 6.3 | 10.0 | 20.0 | 24.5 | 36.5 | 57.3 |

*Nitrite concentrations in mg/kg.

With the exception of the 156 mg/kg of nitrite product, all of the samples exhibited a rapid decrease in nitrite concentration immediately following the curing process (Week 0) and the residual nitrite level continued to steadily decrease over the three week storage period. Hill et al. (1973) observed a similar phenomenon during storage of a variety of cured meats--especially products with high pH (>5.9) and a high moisture content. They suggested that the observed decreases in residual nitrite were due to oxidation-reduction reactions which converted nitrite to nitrous oxide. Much of this undetected nitrite is now believed to react with nonheme protein, low molecular weight peptides and amino acids (Sebranek et al., 1973; Cassens et al., 1974; Kubberod et al., 1974; Goutefongea et al., 1977; Woolford and Cassens, 1977).

It is interesting to note that turkey bologna prepared without sodium nitrite (0 mg/kg) contained detectable levels of residual nitrite. Other researchers have made the same observation (Christiansen et al., 1973; Simon et al., 1973; Gray et al., 1979; Sofos et al., 1979c) and concluded that the nitrite may be from water, spices or other components added during product formulation; or that endogenous substance(s) in meat react as nitrite during the analysis. Hustad et al. (1973) noted that oxides of nitrogen are produced during the smoking procedure and that those compounds can penetrate the meat surface. Although smoke was not used during the cooking of the turkey bologna, it could have been that oxides of nitrogen were still present in the smokehouse and contaminated the product.

d. Hunter colorimetry: The color of turkey bologna prepared with various nitrite levels was objectively analyzed for the L (lightness),

$a_L$ (redness), and $b_L$ (yellowness) values. Analysis of variance (Appendix Table A-13) indicates that time (A), sample treatment (B) and the time-treatment interaction (A X B) were significant ($p < 0.01$) for all three parameters. Due to their interaction, time and treatment effects were not independent and the comparison of treatment means (Appendix Table A-14) was done within each time period. The use of even 20 mg/kg of nitrite increased the redness when compared to a nonnitrite treated sample. However, the introduction of at least 40 mg/kg of nitrite was necessary to provide Hunter color values that were not significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite) over the entire three week storage period. The pronounced effect of nitrite on the Hunter color values has been reported for cured turkey frankfurters (Sales et al., 1980), chicken frankfurters (Gray et al., 1979) and beef-pork frankfurters (MacNeil and Mast, 1973). Although many studies have observed color fading, i.e., decreased $a_L$ values and increased $b_L$ values, during the storage of cured meat products (Froning et al., 1971; Dhillon and Mauer, 1975; Gray et al., 1979) the Hunter color scores in this study remained fairly stable over the three week period at 4°C. Undoubtedly, vacuum packaging the bologna samples aided color stability by reducing the chance of oxidative reactions. Younathan and Watts (1959) and Greene and Price (1975) have concluded that the free radicals produced from oxidizing lipids can oxidize and decompose heme pigments, resulting in an unaesthetic brown color.

The significant relationships ($p < 0.001$) between the Hunter color values and subjective color measurements are indicated in Table 9.

Table 9: Relationship between Hunter color values and subjective evaluation of turkey bologna color.

| Contrast | Regression | Correlation |
|---|---|---|
| Color VS Hunter (L) | Y = 63.50 = 0.23 X | - 0.90 *** |
| Color VS Hunter (a) | Y = 3.30 + 0.32 X | + 0.88 *** |
| Color VS Hunter (b) | Y = 8.90 - 0.26 X | - 0.95 *** |

*** ($p < 0.001$)

It, therefore, appears that Hunter Colorimetry is a useful index for color measurement of cured meat systems.

e. TBA values: The lipid oxidation of turkey bologna during storage was analyzed by the TBA method. Analysis of variance (Appendix Table A-15) indicates that the time (A), sample treatment (B) and the time-treatment interaction (A X B) were significant ($p < 0.01$) effects for this test. Comparison of treatment means within each time interval (Appendix Table A-16) revealed that only the nonnitrite treated samples were significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite). The ability of nitrite to reduce the extent of oxidation compared to samples prepared without sodium nitrite has been demonstrated in chicken frankfurters (Gray et al., 1979), beef-pork frankfurters (MacNeil and Mast, 1973), comminuted cured pork (Greene and Price, 1975; Hadden et al., 1975), and in nitrite treated beef, chicken and pork meat systems (Fooladi et al., 1979; Igene et al., 1979). It was observed that as the level of nitrite introduced into turkey bologna was increased, the TBA values subsequently decreased. A similar relationship between nitrite

concentration and increased antioxidant activity was pointed out by Gray et al. (1979).

Over the three week storage period, the TBA values for turkey bologna prepared without sodium nitrite steadily increased, while the values remained fairly stable for all nitrite treated samples. These findings concur with the reports of Gray et al. (1979) on chicken frankfurters, Dhillon and Mauer (1975) on chicken/turkey summer sausage, MacNeil and Mast (1973) on beef-pork frankfurters and Hadden et al. (1975) on comminuted cured pork.

Several factors must be considered when the antioxidant effectiveness of nitrite is evaluated. Both Fooladi et al. (1979) and Igene et al. (1979) demonstrated that the degree to which nitrite reduced TBA values depended on the meat source. Nitrite was more effective in delaying lipid oxidation in beef or pork than in turkey or chicken. Wilson et al. (1976) reported that red muscles had consistently higher TBA values than white muscle fibers. They concluded that such species differences can be attributed to the type and concentration of lipid present in the meat. Furthermore, Lin and Sebranek (1979) observed that maximum (687-737 mm Hg) initial vacuum levels combined with films of low oxygen permeability (7.0 $ml/m^2$/24 hours or less) aided the antioxidant efficacy of nitrite.

The significant correlations ($p < 0.001$) between TBA values and subjective measurements of off-odor, odor, flavor and overall acceptability are listed in Table 10. This indicates that the undesirable organoleptic characteristics of the bologna were due to lipid oxidation rather than due to microbial action. This is supported by the observation

that sensory panelists frequently detected oxidized and/or rancid flavors and odors in treatment samples which subsequently exhibited higher TBA values. Other studies have confirmed the relationship between TBA values and off-flavor/off-odor detection (Younathan and Watts, 1959; Wilson et al., 1976; Fooladi et al., 1979; Igene et al., 1979).

Table 10: Relationship between TBA values and subjective flavor and odor evaluations of turkey bologna.

| Contrast | Regression | Correlation |
|---|---|---|
| TBA VS Off-Flavor | Y = 2.50 + 0.26 X | + 0.36 ** |
| TBA VS Off-Odor | Y = 0.30 + 0.98 X | + 0.92 *** |
| TBA VS Flavor | Y = 10.54 - 1.11 X | - 0.93 *** |
| TBA VS Odor | Y = 11.30 - 1.11 X | - 0.94 *** |
| TBA VS Acceptability | Y = 12.10 - 1.37 X | - 0.93 *** |

** ($p < 0.05$)

*** ($p < 0.001$)

It should be noted that the correlation between TBA values and the sensory scores of off-flavor was less significant ($p < 0.05$) than the others due to the inconsistency between the subjective scores and the nitrite level in the sample. This was previously mentioned in the discussion of the correlations between the mean panel scores of various sensory attributes for turkey bologna.

f. N-Nitrosamine analysis: Table 11 lists the presumptive results from the N-nitrosamine analysis of turkey bologna cured with various levels of sodium nitrite. Except for trace levels of NDMA in samples treated with 40 and 156 mg/kg of nitrite, only the turkey bologna

Table 11: Presumptive N-nitrosamine levels ( μg/kg) in turkey bologna prepared with various levels of sodium nitrite.

| Nitrite (mg/kg) | NDMA | NDEA | NDBA | NPIP | NPYR | NMOR |
|---|---|---|---|---|---|---|
| 00 | ND* | ND | ND | ND | ND | ND |
| 20 | ND | ND | ND | ND | ND | ND |
| 40 | TR** | ND | ND | ND | ND | ND |
| 60 | ND | ND | ND | ND | ND | ND |
| 100 | ND | ND | ND | ND | ND | ND |
| 156 | TR | ND | ND | ND | 0.5 | 0.5 |

*ND, none detected, less than the limit of detection.
**TR, trace levels ( < 0.3 μg/kg).

prepared with the highest level of nitrite (156 mg/kg) contained measurable levels of N-nitrosamines. These samples contained 0.5 μg/kg of both NPYR and NMOR. These results agree with the N-nitrosamine analyses of canned, comminuted cured ham (Christiansen et al., 1973), beef-pork frankfurters (Hustad et al., 1973) and thuringer sausage (Dethmers and Rock, 1975). Even when these products were manufactured with 300 mg/kg of sodium nitrite, the analyses for volatile N-nitrosamines were still negative. It should be noted, however, that the detection limit for those studies was only 10 μg/kg, while the GC-TEA used in this turkey bologna project was much more sensitive ( 0.5 μg/kg). Many investigators have researched the precursors and mechanism for NPYR formation (Lijinsky and Epstein, 1970); Ender and Ceh, 1971; Bills et al., 1973; Gray and Dugan, 1975; Gray and Collins, 1977, 1978; Bharucha et al., 1979), but the source of NMOR has not been studied. A discussion on NMOR formation will follow in the N-nitrosamine analysis section for nitrite treated turkey hams.

The sensory and chemical analyses of turkey bologna chubs prepared with various levels of sodium nitrite indicated that the introduction of approximately 40 mg/kg of nitrite provided a product that was organoleptically acceptable, exhibited reduced TBA values and had Hunter color results which were not significantly ($p < 0.01$) different from the control (156 mg/kg of nitrite). Therefore, the levels of 40 and 60 mg/kg nitrite were selected for use in Test 2, to be incorporated alone or in combination with potassium sorbate.

2. Test 2: Nitrite-Sorbate Combinations

a. Organoleptic analysis: Turkey bologna chubs prepared with various levels of sodium nitrite and potassium sorbate (alone or in combination) were evaluated at regular intervals by a sensory panel for color, odor, flavor, off-odor, off-flavor and overall acceptability. The influence of nitrite-sorbate treatments on the mean panel scores for color is shown in Figure 8. Analysis of variance (Appendix Table A-17) indicated that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-18) revealed that the nonnitrite treated samples and those containing only sorbate (0.26 and 0.39%) were significantly different ($p < 0.01$) from the reference. Evidently, sorbate has the same disadvantage as many other potential nitrite substitutes, in that it cannot emulate the effect of nitrite on cured meat color. However, when sorbate was combined with reduced levels of nitrite, the resulting color of the turkey bologna was not significantly different ($p < 0.01$) from the reference.

Figure 8: Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna color during storage at 4°C.

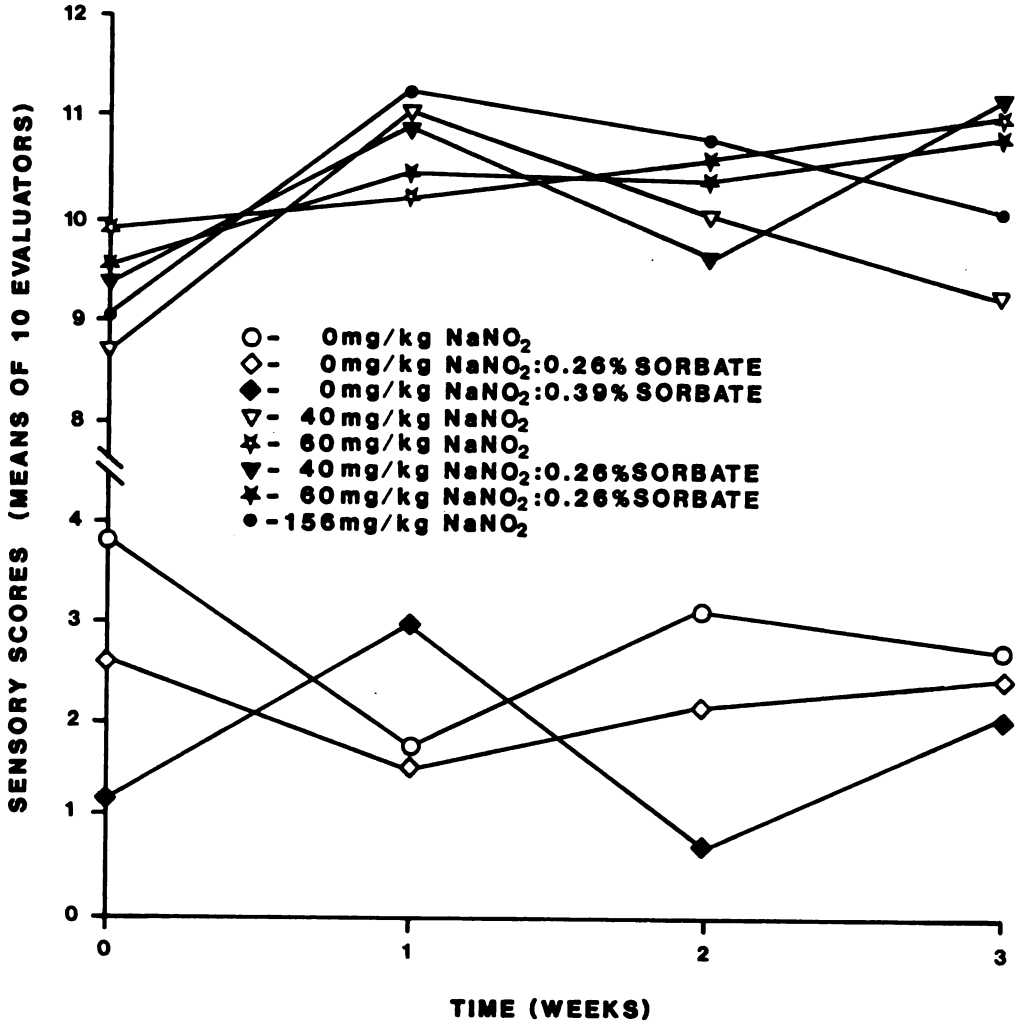

SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
8
9
10
11
12
○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✡- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2

Figure 8

As the level of nitrite added to the turkey bologna increased, the color acceptability of the treatment sample increased, regardless of whether the nitrite was introduced alone or in combination with 0.26% sorbate. It was observed, however, that the samples containing both nitrite and sorbate exhibited lower color scores than the corresponding product prepared with nitrite alone. Furthermore, the turkey bologna manufactured with only sorbate (0.26 or 0.39%) received lower color scores than the product prepared without sodium nitrite. However, both of these effects varied over time and so they were attributed to product variation. Gray et al. (1979) reported similar results for chicken frankfurters.

The influence of nitrite-sorbate combinations on the mean sensory scores for turkey bologna flavor and odor is shown in Figures 9 and 10, respectively. Analysis of variance (Appendix Tables A-19 and A-20) indicates that only sample treatment (B) was significant ($p < 0.01$) for both attributes. Comparison of treatment means for flavor and aroma scores (Appendix Tables A-21 and A-22, respectively) reveals that samples prepared without nitrite (0 mg/kg) or with sorbate alone (0.26 or 0.39%) were significantly different ($p < 0.01$) from the reference (156 mg/kg of nitrite). Obviously, sorbate itself does not contribute to the characteristic flavor of cured meats, but when 0.26% sorbate was combined with reduced levels of nitrite (40 to 60 mg/kg), the bologna exhibited a flavor comparable to the sample prepared with 156 mg/kg of nitrite. Paquette et al. (1980) reported similar results for bacon processed with 0.26% potassium sorbate in combination with 40 or 80 mg/kg of sodium nitrite.

Figure 9: Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna flavor during storage at 4°C.

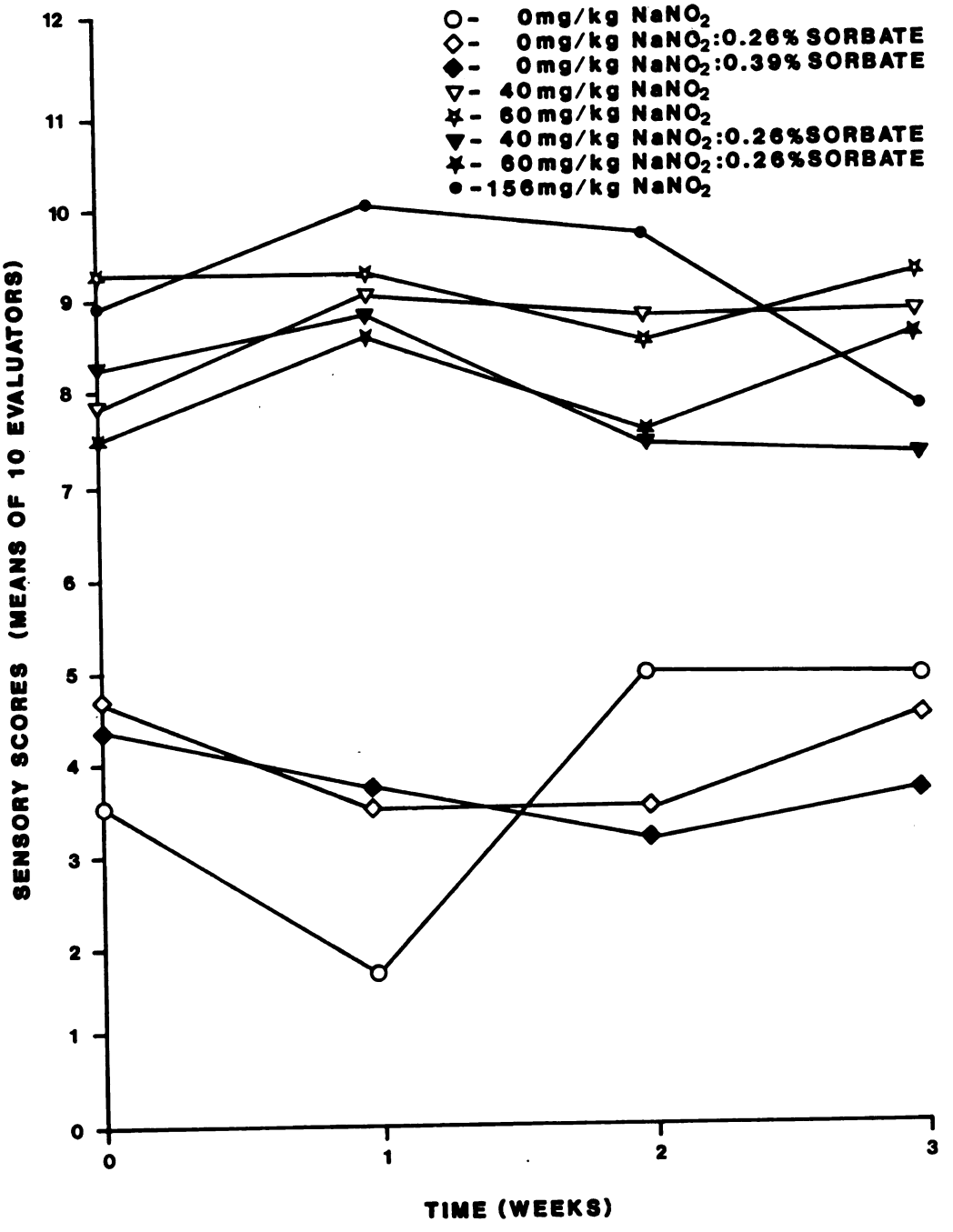

○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✡- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
0
1
2
3
4
5
6
7
8
9
10
11
12

Figure 9

Figure 10: Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna odor during storage at 4°C.

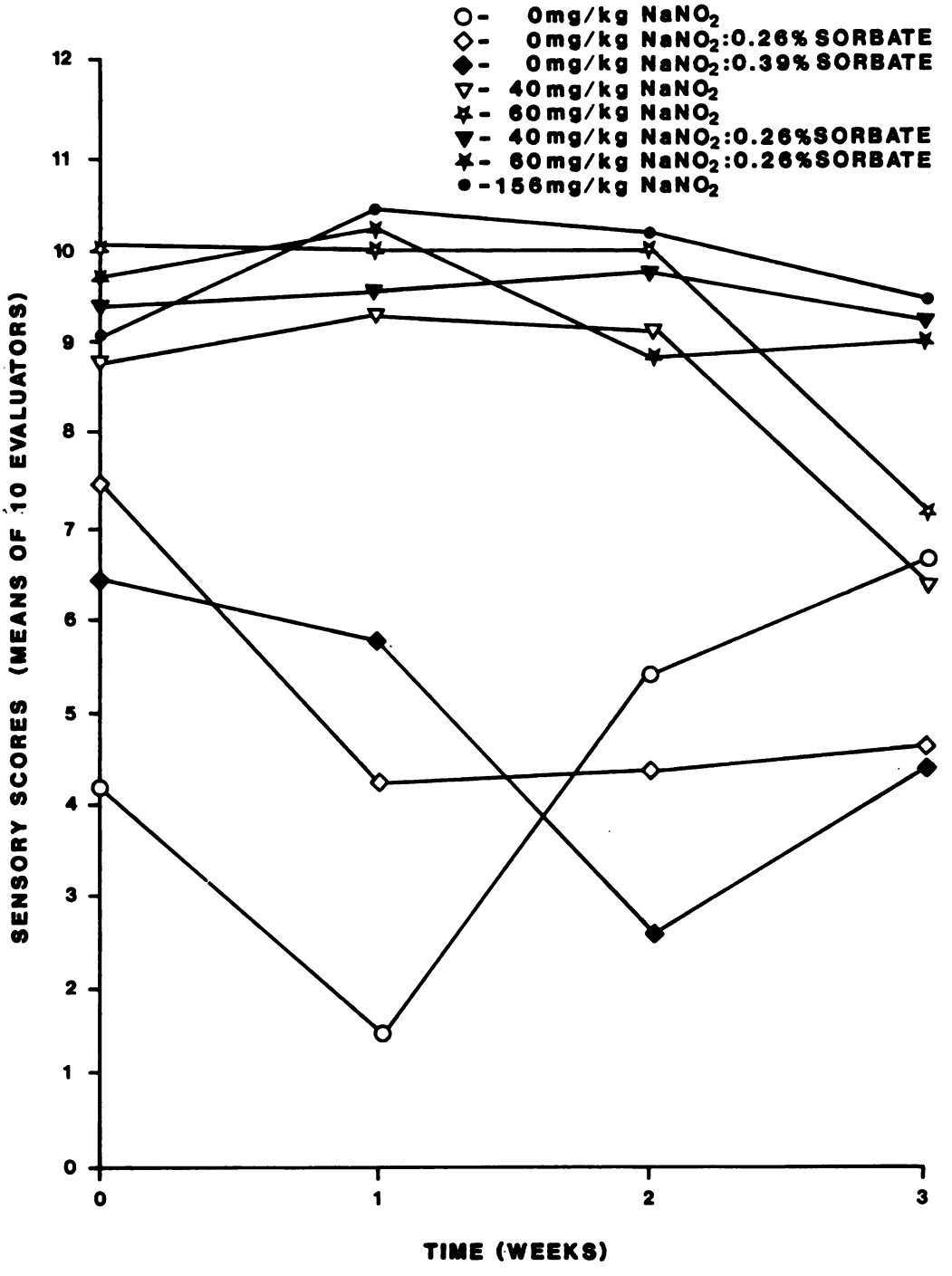

○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✲- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✲- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
0
1
2
3
4
5
6
7
8
9
10
11
12
TIME (WEEKS)

Figure 10

For the flavor attribute, increased nitrite input resulted in increased scores. However, products containing nitrite in combination with sorbate exhibited lower flavor scores than the corresponding samples containing nitrite alone. This was reflected in the panelists comments that the addition of sorbate contributed a sweet,"saccharin-like" flavor to the product. This flavor defect was readily detected in bologna prepared with sorbate alone, but the presence of nitrite decreased perception to a slight extent. Apparently, nitrite addition increased the cured flavor of the product and thereby masked the flavor contribution from sorbate.

For turkey bologna aroma, increased nitrite input resulted in increased sensory scores. Products prepared with reduced levels of nitrite (either alone or in combination with sorbate) were comparable to the reference and the differences between the various treatment groups were attributed to product variation.

The influence of nitrite-sorbate treatments on the mean sensory scores for turkey bologna off-flavor and off-odor is shown in Figures 11 and 12, respectively. Analysis of variance (Appendix Tables A-23 and A-24) reveals that only sample treatment (B) exerted a significant ($p < 0.01$) effect on both attributes. For both off-flavor and off-odor scores, comparison of treatment means over time (Appendix Tables A-25 and A-26) indicates that the incorporation of at least 40 mg/kg of nitrite, alone or combined with 0.26% sorbate, provided a product that was not significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite). As mentioned in the discussion of turkey bologna flavor, panelists detected a sweet, "saccharin-like" flavor along with a

Figure 11: Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna off-flavor during storage at 4°C.

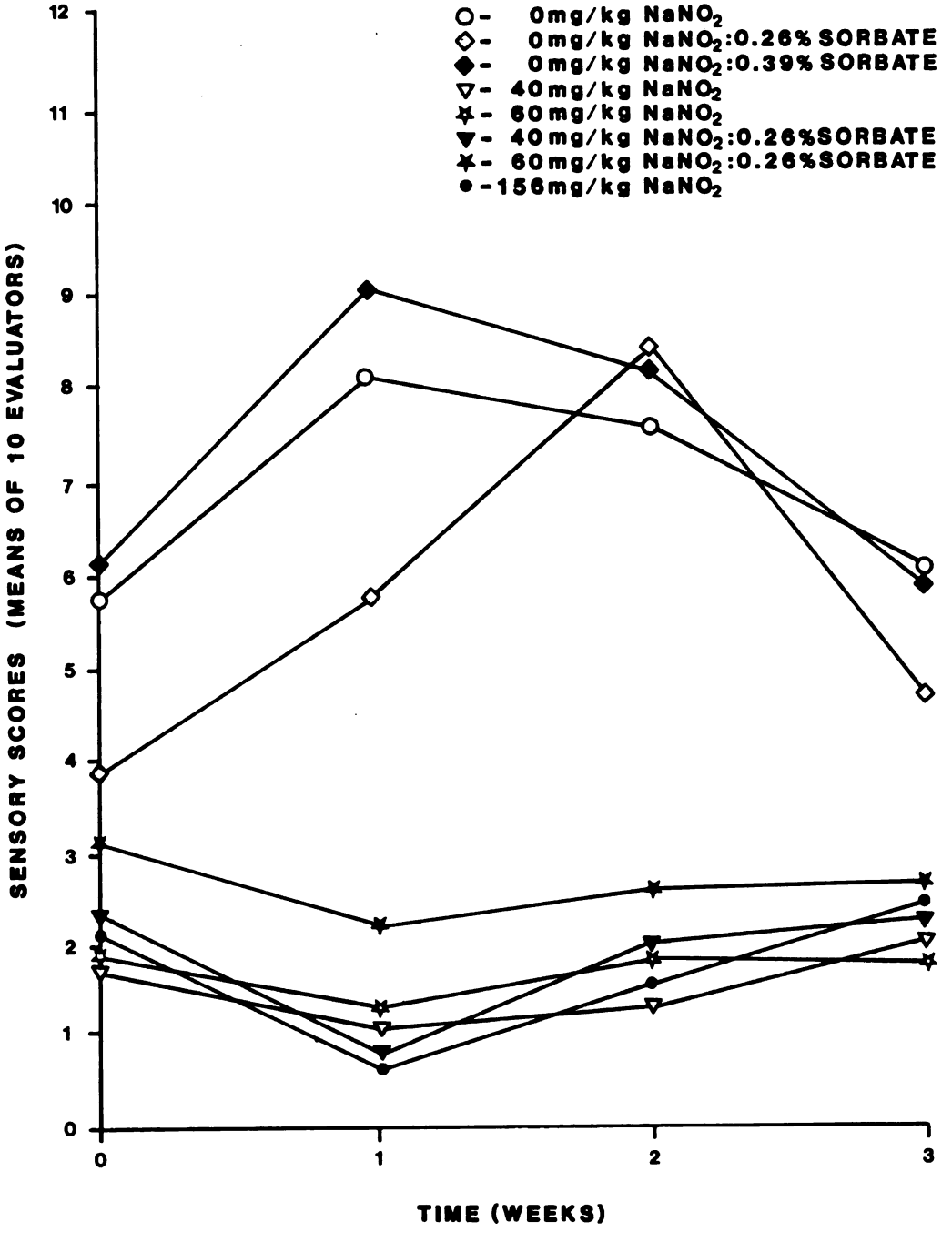

○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✲- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
0
1
2
3
4
5
6
7
8
9
10
11
12
0
1
2
3
TIME (WEEKS)

Figure 11

Figure 12: Effect of various nitrite-sorbate treatments on the mean panel scores for turkey bologna off-odor during storage at 4°C.

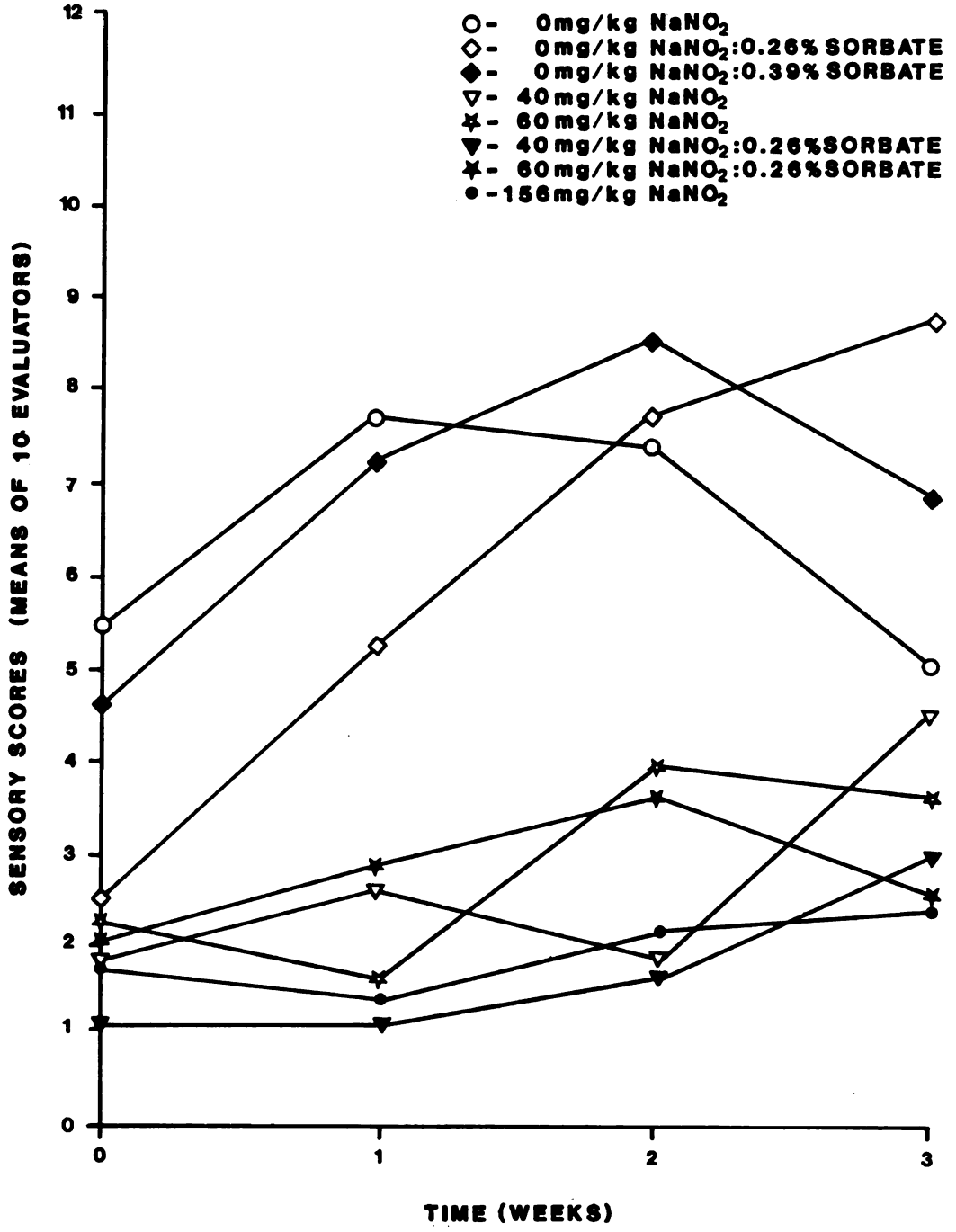

○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✱- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
0
1
2
3
4
5
6
7
8
9
10
11
12
0
1
2
3
TIME (WEEKS)

Figure 12

bitter/oxidized aftertaste in samples prepared with sorbate. Apparently, the panel members considered these contributions from sorbate to be "off" or undesirable because turkey bologna containing nitrite-sorbate combinations received greater off-flavor scores than the corresponding samples prepared with nitrite alone. Other authors have reported the existence of sweet, "chemical-like" and/or sweet aromatic flavors and aromas in meat products manufactured with sorbate, either alone or combined with reduced levels of nitrite (USDA, 1979; Berry and Blumer, 1981). Clearly, these flavor acceptance problems are a distinct disadvantage against sorbate use.

Although the subjective evaluation of off-flavor presented some general trends, the organoleptic results for off-odor were inconsistent. For bologna prepared with sorbate alone, the off-odor scores increased with increased sorbate level. However, the combination of 40 mg/kg of nitrite and 0.26% sorbate was considered to have less detectable off-odor than even the reference sample, and the bologna prepared with 60 mg/kg of nitrite and 0.26% sorbate received greater off-odor values than any of the samples containing nitrite (alone or combined with sorbate).

The influence of various nitrite-sorbate combinations on the mean sensory scores for the overall acceptability of turkey bologna is shown in Figure 13. Analysis of variance (Appendix Table A-27) indicates that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-28) reveals that nonnitrite treated samples and products containing only sorbate (0.26 or 0.39%) were significantly different ($p < 0.01$) from

Figure 13: Effect of various nitrite-sorbate treatments on the mean panel scores for the overall acceptability of turkey bologna during storage at 4°C.

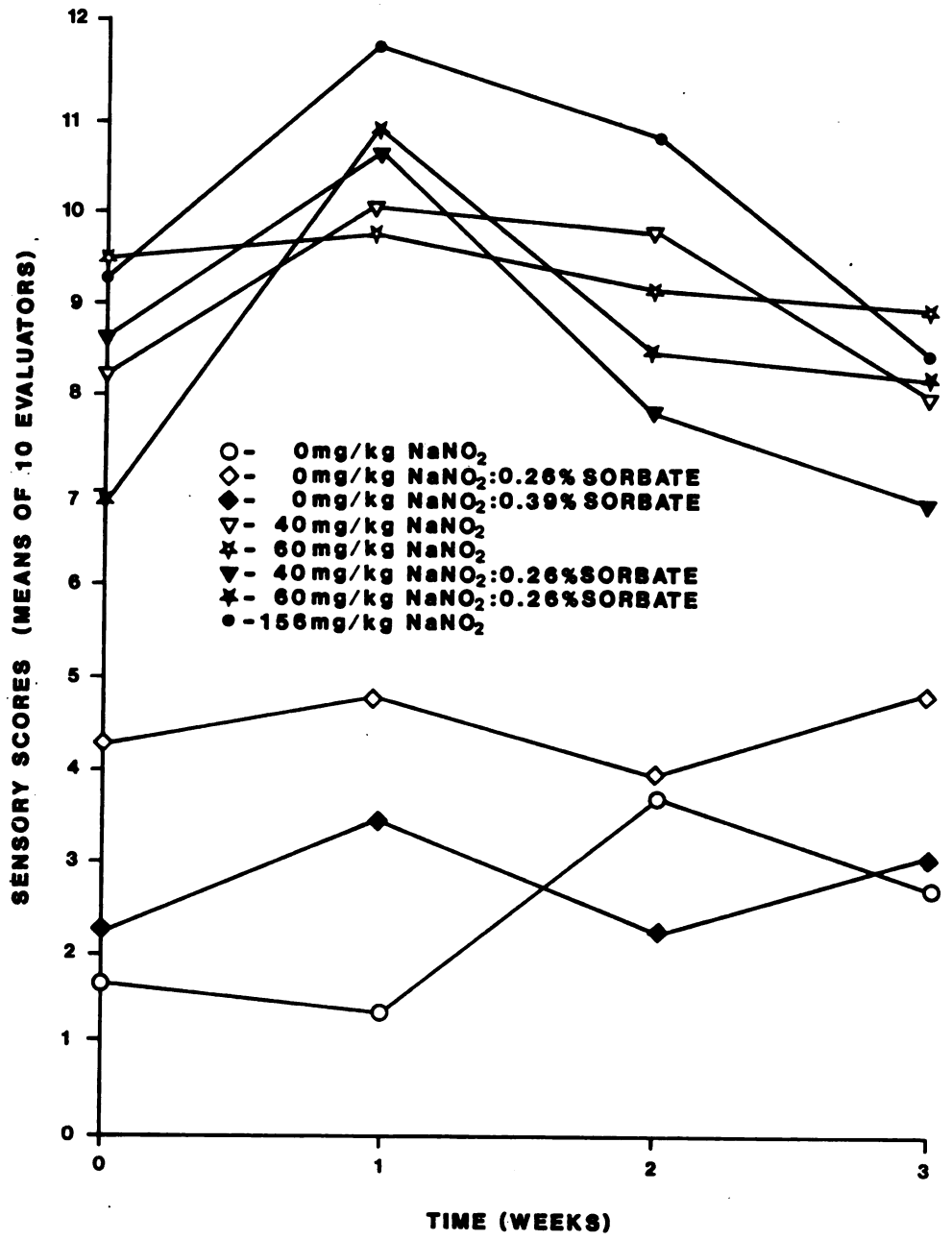

SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
6
7
8
9
10
11
12
○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✡- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2

Figure 13

the control (156 mg/kg of nitrite). Increased nitrite input or the combination of nitrite and sorbate in the bologna product did not provide any consistent trends in the sensory scores for overall acceptability. However, all such treatment groups were comparable to the control. Similar results have been reported for chicken frankfurters (Gray et al., 1979) and bacon (Paquette et al., 1980). It also appeared that color was the major factor in the acceptance of a product. Many panelists commented that bologna prepared without nitrite (0 mg/kg) or with only sorbate (0.26 or 0.39%) would have been more acceptable had the color been more aesthetically pleasing. Red or green lights were not used to mask the color differences between treatment groups. So, a brown/undesirable hue in a product may have influenced the scores subsequently assigned to the other sensory attributes of the same sample.

The linear correlation and linear regression analyses conducted among the various sensory attributes of turkey bologna are listed in Table 12. Increased bologna aroma and flavor were associated with decreased off-odor and off-flavor effects. Also, increased flavor/aroma intensity or decreased off-flavor/off-odor intensity was significantly ($p < 0.001$) correlated with greater product acceptability.

b. Proximate analysis and pH: Table 13 lists the moisture, fat and protein analyses for raw turkey thigh meat and for turkey bologna prepared without sodium nitrite (0 mg/kg). The fat content was closer to the 25% target level than the product in Test 1. These results are similar to those reported for chicken frankfurter emulsions (Sofos et al., 1979c) and for canned, comminuted pork (Ivey and Robach, 1978).

Table 12: Linear correlation and linear regression of various sensory attributes for turkey bologna containing nitrite-sorbate treatments.

| Contrast | | Regression | Correlation |
|---|---|---|---|
| Flavor | VS Acceptability | Y = -1.39 + 1.22 X | + 0.95 *** |
| Color | VS Acceptability | Y = 1.54 + 0.73 X | + 0.92 *** |
| Odor | VS Acceptability | Y = -1.11 + 1.10 X | + 0.86 *** |
| Off-Flavor | VS Acceptability | Y = 10.81 - 1.21 X | - 0.91 *** |
| Off-Odor | VS Acceptability | Y = 10.91 - 1.07 X | - 0.80 *** |
| Off-Flavor | VS Off-Odor | Y = 0.90 + 0.82 X | + 0.88 *** |
| Off-Flavor | VS Flavor | Y = 9.95 - 0.91 X | - 0.92 *** |
| Off-Flavor | VS Odor | Y = 10.58 - 0.86 X | - 0.90 *** |
| Off-Odor | VS Odor | Y = 11.24 - 0.97 X | - 0.90 *** |
| Off-Odor | VS Flavor | Y = 10.09 - 0.88 X | - 0.83 *** |
| Odor | VS Flavor | Y = 0.21 + 0.87 X | + 0.89 *** |

*** ($p < 0.001$)

Table 13: Proximate analysis of raw turkey thigh meat and nonnitrite treated turkey bologna.

| Sample | Moisture (%) | Fat (%) | Protein (%) |
|---|---|---|---|
| Raw meat | 76.17 ± 0.32 | 3.58 ± 0.38 | 17.55 ± 0.43 |
| Turkey bologna | 59.69 ± 0.28 | 21.66 ± 0.98 | 12.77 ± 0.54 |

The pH values for the turkey bologna samples (Table 14) are comparable to those reported for cooked, uncured sausages (Tompkin et al., 1974) and chicken frankfurters (Grey et al., 1979), but are more alkaline than the results reported for chicken/turkey frankfurters (Huhtanen and Feinberg, 1980). This is probably because no acidulants were added to the turkey bologna. The pH readings remained fairly stable

Table 14: pH values for turkey bologna prepared with various nitrite-sorbate treatments and stored at 4°C.

| Treatment | Time (weeks) | | | | |
|---|---|---|---|---|---|
| | 0 | 1 | 2 | 3 | X |
| 00* | 6.37 | 6.18 | 6.25 | 6.28 | 6.27 ± .08 |
| 02 | 6.30 | 6.23 | 6.30 | 6.33 | 6.29 ± .04 |
| 03 | 6.30 | 6.25 | 6.30 | 6.33 | 6.29 ± .03 |
| 40 | 6.34 | 6.20 | 6.28 | 6.28 | 6.29 ± .06 |
| 60 | 6.30 | 6.18 | 6.25 | 6.30 | 6.26 ± .06 |
| 42 | 6.36 | 6.20 | 6.30 | 6.33 | 6.29 ± .07 |
| 62 | 6.32 | 6.23 | 6.30 | 6.35 | 6.30 ± .05 |
| 156 | 6.32 | 6.23 | 6.30 | 6.33 | 6.29 ± .05 |

*See Table 3 for nitrite-sorbate treatments corresponding to these identification numbers.

over the three week storage for all treatment groups. Similarly, Gray et al. (1979) reported that chicken frankfurters prepared with various nitrite-sorbate combinations exhibited little pH change during refrigerated storage. It is interesting to note that the turkey bologna exhibited little/no pH change when increased sodium nitrite was incorporated or when potassium sorbate was added. Although sorbic acid has been reported to have an acidifying effect on many meat products (Tompkin et al., 1974; Ivey and Robach, 1978; Gray et al., 1979), the salt form, potassium sorbate, does not appear to influence pH to any extent (Huhtanen and Feinberg, 1980).

c. Residual nitrite: The residual nitrite analysis of turkey bologna prepared with various nitrite-sorbate combinations is listed in Table 15. As was found in Test 1, increased nitrite input resulted in greater residual nitrite, the level of residual nitrite decreased over

time for all treatments, and detectable nitrite was found in nonnitrite treated samples. These results are consistent with other reports on chicken frankfurter emulsions (Sofos et al., 1979c, 1980a) and canned comminuted pork (Ivey and Robach, 1978; Lee et al., 1978).

Table 15: Residual nitrite analysis of turkey bologna prepared with various nitrite-sorbate combinations and stored at 4°C.

| Time (weeks) | Nitrite-Sorbate Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00* | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| Week 0 | 3.5** | 3.8 | 2.8 | 22.8 | 32.3 | 18.3 | 36.3 | 87.8 |
| Week 1 | 2.8 | 2.3 | 2.5 | 17.8 | 27.0 | 15.8 | 32.0 | 75.0 |
| Week 2 | 2.5 | 2.5 | 2.5 | 18.1 | 25.6 | 15.9 | 30.5 | 72.1 |
| Week 3 | 4.5 | 4.0 | 4.3 | 19.5 | 24.3 | 15.3 | 29.3 | 70.8 |

*See Table 3 for nitrite-sorbate treatments corresponding to these identification numbers.

**Nitrite concentrations in mg/kg.

Previous research (Sofos et al., 1979c, 1980a) reported that the presence of sorbate decreased the rate of nitrite depletion and/or resulted in a higher residual nitrite levels during storage. These facts are supported by comparison of residual nitrite levels in turkey bologna prepared with 60 mg/kg of nitrite alone and the samples containing 60 mg/kg of nitrite combined with 0.26% sorbate. The reason(s) for such an effect is unknown, but it has been shown to be pH dependent. Sofos et al. (1980a) compared chicken frankfurter emulsions at two pH values (6.20 and 7.15) and observed that at the lower product pH level (6.20) residual nitrite depletion was slower. They commented that such a finding was unexpected since at lower pH values nitrite would be

expected to form nitrous acid and disappear more rapidly than at high pH values.

d. Hunter colorimetry: The color of turkey bologna processed with various nitrite-sorbate combinations was objectively analyzed for the L (lightness), $a_L$ (redness) and $b_L$ (yellowness) values. Analysis of variance (Appendix Table A-29) indicated that time (A), treatment (B) and the time-treatment interaction (A X B) were significant ($p < 0.01$) for all three parameters. Comparison of treatment means within each time period (Appendix Table A-30) showed that nonnitrite treated samples and those prepared with sorbate alone (0.26 and 0.39%) were significantly different ($p < 0.01$) from the control for the $a_L$ and $b_L$ values. Such an effect was not consistently shown for product lightness (L). The presence of nitrite, alone or in combination with sorbate, resulted in a darker, redder and less yellow product than when no nitrite or only sorbate was added. Similar results have been reported for chicken frankfurters prepared with various nitrite-sorbate treatments (Gray et al., 1979).

Increased nitrite input had little effect on the L, $a_L$ or $b_L$ values, but the combination of sorbate with reduced levels of nitrite (40 or 60 mg/kg) provided less redness and more yellowness than was observed in the corresponding product containing nitrite alone. This corresponds with the subjective evaluation of bologna color, where samples containing both sorbate and nitrite received lower color scores than their counterparts processed with only nitrite. Over the three week storage period, L and $b_L$ values remained fairly stable for all treatment groups, while the $a_L$ values gradually decreased.

Linear correlation and linear regression analyses between subjective and objective measurements of bologna color are listed in Table 16. The significant ($p < 0.001$) relationships indicate that Hunter color values are useful indices for color measurement of cured meat systems.

Table 16: Relationship between Hunter color values and the subjective evaluation of turkey bologna color.

| Contrast | Regression | Correlation |
|---|---|---|
| Color VS Hunter (L) | Y = 66.24 - 0.07 X | - 0.63 *** |
| Color VS Hunter (a) | Y = 2.63 + 0.31 X | + 0.94 *** |
| Color VS Hunter (b) | Y = 10.03 - 0.25 X | - 0.97 *** |

***($p < 0.001$)

e. TBA values: The lipid oxidation in turkey bologna stored over a three week period at 4°C was objectively analyzed by the TBA method. Analysis of variance (Appendix Table A-31) showed that time (A), treatment (B) and the time-treatment interaction (A X B) were all significant ($p < 0.01$) for this test. Comparison of treatment means within each time interval (Appendix Table A-32) revealed that the absence of nitrite or the addition of sorbate alone (0.26 or 0.39%) resulted in a significantly ($p < 0.01$) greater amount of lipid oxidation in the turkey bologna. Evidently, sorbate itself has no antioxidant activity because when 40 mg/kg or more of nitrite was incorporated into the cure formulation (alone or combined with 0.26% sorbate) the resulting product exhibited reduced TBA values which were not significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite).

Increased nitrite input decreased the degree of lipid oxidation in the turkey bologna. Similar results were reported for chicken frankfurters by Gray et al. (1979). They also observed that the presence of sorbate did not influence the TBA results. In contrast, the study reported here found that turkey bologna prepared with nitrite and sorbate combinations exhibited greater TBA values than the corresponding product containing only nitrite. Such results are surprising because sorbate has been shown to slow the rate of residual nitrite depletion, and therefore one would expect less lipid oxidation (lower TBA values) in products prepared with both nitrite and sorbate.

Over the storage period, the TBA values appeared to increase for both the nonnitrite treated samples and for those prepared with sorbate alone. However, for samples containing nitrite, alone or in combination with 0.26% sorbate, the results were variable and did not provide any consistent trends.

The significant ($p < 0.01$) correlations between TBA values and the subjective evaluations of odor, flavor, off-odor, off-flavor and acceptability are listed in Table 17.

Table 17: Relationship between TBA values and subjective flavor and odor evaluations for turkey bologna.

| Contrast | Regression | Correlation |
|---|---|---|
| TBA VS Off-Flavor | Y = 0.49 + 0.79 X | + 0.91 *** |
| TBA VS Off-Odor | Y = 1.08 + 0.71 X | + 0.89 *** |
| TBA VS Flavor | Y = 0.76 - 0.78 X | - 0.93 *** |
| TBA VS Odor | Y = 10.49 - 0.77 X | - 0.89 *** |
| TBA VS Acceptability | Y = 10.42 - 0.93 X | - 0.87 *** |

*** ($p < 0.001$)

This indicates that the undesirable flavor and odor characteristics of the bologna could be attributed to lipid oxidation rather than due to microbial action.

f. N-Nitrosamine analysis: Turkey bologna chubs prepared with various nitrite-sorbate treatments were analyzed for the presence of volatile N-nitrosamines using the GC-TEA system. The presumptive results are listed in Table 18. Samples processed with 0.39% sorbate alone, and with 40 and 156 mg/kg of nitrite contained trace levels of NDMA. Only the bologna cured with the highest level of sodium nitrite (156 mg/kg) contained N-nitrosamines at a level greater than 0.3 µg/kg. This treatment group contained 0.5 µg/kg of both NPYR and NMOR. The fact that meat products processed with potassium sorbate and reduced levels of sodium nitrite exhibit low or zero levels of volatile N-nitrosamines has been reported by other researchers (Ivey et al., 1978; Robach et al., 1980c).

Table 18: Presumptive N-nitrosamine levels (µg/kg) in turkey bologna prepared with various nitrite-sorbate treatments.

| Treatment | NDMA | NDEA | NDBA | NPIP | NPYR | NMOR |
|---|---|---|---|---|---|---|
| 00[a] | ND[b] | ND | ND | ND | ND | ND |
| 02 | ND | ND | ND | ND | ND | ND |
| 03 | TR[c] | ND | ND | ND | ND | ND |
| 40 | TR | ND | ND | ND | ND | ND |
| 60 | ND | ND | ND | ND | ND | ND |
| 42 | ND | ND | ND | ND | ND | ND |
| 62 | ND | ND | ND | ND | ND | ND |
| 156 | TR | ND | ND | ND | 0.5 | 0.5 |

[a]See Table 3 for the nitrite-sorbate treatments corresponding to these identification numbers.

[b]ND, none detected, less than the limit of detection.

[c]TR, trace levels (0.3 µg/kg).

g. Clostridium botulinum study: The efficacy of various nitrite-sorbate combinations in inhibiting C. botulinum growth and toxinogenesis is demonsttrated in Table 19. After five days of temperature abuse at 27°C, turkey bologna prepared without sodium nitrite or with only potassium sorbate (0.26 or 0.39%) had sufficient toxin formation to cause the death of all injected mice. The introduction of 40 mg/kg of nitrite, alone or in combination with 0.26% sorbate, delayed toxin development until the end of one week of incubation, but inoculated samples prepared with 60 mg/kg of nitrite alone did not exhibit toxin production until after four weeks at 27°C. The combination of 60 mg/kg of nitrite and 0.26% sorbate appeared to be as effective as 156 mg/kg of nitrite alone in inhibiting C. botulinum over the entire four week storage period.

Similar to the results of this study, researchers have reported that nitrite-free meat samples, when inoculated with C. botulinum spores, developed toxin within 4 to 6 days at 27°C, and that as the level of nitrite increased, the probability of botulinal toxin production decreased (Christiansen et al., 1973; Ivey and Robach, 1978; Sofos et al., 1979c). Although some investigators argue that 50 mg/kg of nitrite is sufficient to inhibit C. botulinum (Hustad et al., 1973; Bowen et al., 1974), others contend that greater nitrite concentrations (100 to 156 mg/kg) are required (Gray et al., 1979; Sofos et al., 1979c).

The incorporation of sorbate into cure formulations has reduced the levels of nitrite necessary to insure the botulinal safety of the product. This synergistic effect has been shown in the turkey bologna study and has also been reported for chicken frankfurter emulsions (Robach et al., 1978; Gray et al., 1979; Sofos et al., 1979b,c) and

Table 19: Influence of nitrite-sorbate combinations on the *Clostridium botulinum* toxinogenesis in turkey bologna held at 27°C over 28 days.

| Treatment[a] | Time (days) 0 | 3 | 5 | 7 | 10 | 14 | 21 | 28 |
|---|---|---|---|---|---|---|---|---|
| 00-I[b] | -- | -- | 2-D[c] | 2-D | 2-D | 2-D | 2-D | 2-D |
| 00-II | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 00-III | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 02-I | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 02-II | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 02-III | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 03-I | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 03-II | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 03-III | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D | 2-D |
| 40-I | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 40-II | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 40-III | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 60-I | -- | -- | -- | -- | -- | -- | -- | 2-D |
| 60-II | -- | -- | -- | -- | -- | -- | 2-D | 2-D |
| 60-III | -- | -- | -- | -- | -- | -- | -- | 2-D |
| 42-I | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 42-II | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 42-III | -- | -- | -- | 2-D | 2-D | 2-D | 2-D | 2-D |
| 62-I | -- | -- | -- | -- | -- | 2-D | 2-D | -- |
| 62-II | -- | -- | -- | -- | -- | -- | -- | -- |
| 62-III | -- | -- | -- | -- | -- | -- | -- | -- |
| 156-I | -- | -- | -- | -- | -- | -- | -- | -- |
| 156-II | -- | -- | -- | -- | -- | 2-D | -- | -- |
| 156-III | -- | -- | -- | -- | -- | -- | -- | -- |

[a]See Table 3 for the nitrite-sorbate treatments corresponding to these identification numbers.

[b]Three samples per treatment group were analyzed.

[c]2-D, both of the injected mice died after exhibiting the typical symptoms of botulism.

canned comminuted pork (Ivey and Robach, 1978). Several reasons for this synergistic effect have been proposed, including: (1) sorbate delays nitrite depletion, so more nitrite is available to inhibit C. botulinum, (2) the individual effects that sorbate and nitrite exert on C. botulinum become additive when both compounds are present in a system, and (3) nitrite increased the pH range at which sorbate is effective (Sofos et al., 1979c, 1980a). In contrast with the reports by other researchers (Ivey and Robach, 1978; Robach et al., 1978; Gray et al., 1979; Sofos et al., 1979c), the combination of 40 mg/kg of nitrite and 0.26% sorbate used in this study was not as effective as the reference (with 156 mgkg of nitrite). In fact, this combination did not exhibit any more antibotulinal efficacy than when 40 mg/kg of nitrite was used alone. This ineffectiveness could have been due to insufficient residual nitrite and sorbate concentration, or to an insufficient decrease in pH for the sorbate to be effective (Sofos et al., 1979b).

Furthermore, other reports have indicated that 0.26 to 0.39% sorbate alone can be as effective as 156 mg/kg of nitrite in delaying toxin development (Ivey and Robach, 1978; Gray et al., 1979; Sofos et al., 1979c). However, turkey bologna prepared with 0.26 or 0.39% sorbate alone developed toxin as quickly as the sample prepared without nitrite. It is possible that sorbate might have exhibited greater inhibitory activity had the pH of the bologna been lower than 6.20. Sofos et al. (1980a) observed that when sorbate is incorporated into a formulation alone, the pH of the product must be 6.0 or below for sorbate to be effective.

# IV. TURKEY HAM STUDY

## A. EXPERIMENTAL

This study on turkey ham was again divided into two sections. The initial research, henceforth referred to as Test 1, determined the minimum level of sodium nitrite which provided an acceptable turkey ham product according to organoleptic evaluation and chemical analyses. The second half of the study (Test 2) evaluated turkey ham prepared with potassium sorbate, alone or in combination with reduced levels of sodium nitrite, according to organoleptic, chemical and microbiological methods.

### 1. Ham Preparation

The turkey hams for both Tests 1 and 2 were prepared by the following procedures. Deboned turkey thigh pieces were purchased as needed from a commercial processor (Bill-Mar Foods). Formulations were computed to yield an 8% added moisture level in the finished product based on an anticipated 90% smokehouse yield. Appropriate amounts of turkey thigh meat, ice water, spice mix (B. Hellar Company, 8 ounces/100 pounds of meat), sugar (0.73% of the finished product), salt (2.2%), sodium ascorbate (550 mg/kg) and curing phosphate (0.25%, equimixture of sodium tripolyphosphate, sodium pyrophosphate, sodium hexmetaphosphate and monosodium phosphate, Griffith Labs., Chicago, IL.) were introduced into a 80 kg capacity paddle mixer (Smith Company, Buffalo, NY.) and

mixed for 5 minutes without vacuum. Predetermined amounts of sodium nitrite and/or potassium sorbate (Tables 20 and 21) were introduced into the meat mixture and mixed one minute under vacuum to assist the homogeneous distribution of the additives. The temperature of the blends was recorded (range of -1° to 4°C for both tests).

Table 20: Target nitrite levels for turkey ham (Test 1).

| Treatment No. | Sodium Nitrite (mg/kg) |
|---|---|
| 00 | 0 |
| 20 | 20 |
| 40 | 40 |
| 60 | 60 |
| 100 | 100 |
| 156 | 156 |

Table 21: Target nitrite-sorbate levels for turkey ham (Test 2).

| Treatment No. | Sodium Nitrite (mg/kg) | Potassium Sorbate (%) |
|---|---|---|
| 00 | 0 | 0.0 |
| 02 | 0 | 0.26 |
| 03 | 0 | 0.39 |
| 40 | 40 | 0.0 |
| 60 | 60 | 0.0 |
| 42 | 40 | 0.26 |
| 62 | 60 | 0.26 |
| 156 | 156 | 0.0 |

The meat system was transferred to a water pressure sausage stuffer (E.F. Zuber, Minneapolis, MN.) and stuffed into 150 mm cellulose casings.

The approximately 3.5 kg chubs were cooked in a smokehouse equipped with temperature and humidity controls (Drying Systems, Inc., Chicago, IL.). No smoke was introduced into the heating process listed in Table 22. Products were cooked to a minimum internal temperature of 68.8°C (156°F) and cooled for 20 minutes by a cold water shower. The chubs were held at room temperature for two hours before storing overnight at 4°C. Weights of the individual hams were recorded before and after cooking with resultant yields from 88 to 90% for Test 1 and from 88 to 93% for Test 2. The casings were removed and 1 kg ham sections were vacuum packaged (Multivac, West Germany) in Vac 4 ® bags (Koch, Kansas City, MO.) for three weeks storage at 4°C.

Table 22: Cooking schedule for turkey hams (Tests 1 and 2).

| Time (minutes) | Dry Bulb (°C) | Wet Bulb (°C) | R.H. (%) |
|---|---|---|---|
| 30 | 54 | 32-35 | 22 |
| 90 | 63 | 41-42 | 32 |
| 90 | 71 | 54-57 | 40-45 |
| 90 | 79 | 68-71 | 60-65 |
| * | 82 | 71-74 | (steam) |

*Time necessary to reach 68.8°C internal temperature.

2. Organoleptic Analysis

Procedures for the sensory evaluation of the turkey hams were similar to those described for the turkey bologna product.

3. Analytical Methods

Procedures for proximate analysis, residual nitrite, TBA, Hunter Colorimetry and pH determinations were the same as those described for the turkey bologna.

4. Statistical Analysis

Please refer to the corresponding sectin on turkey bologna.

5. N-Nitrosamine Study

Procedures were the same as those described for the turkey bologna. Recoveries for all N-nitrosamines average 107% for 2 μg/kg spiking, 99% for 5 μg/kg spiking and 96% for 10 μg/kg spiking. Since only the reference sample (prepared with 156 mg/kg sodium nitrite) yielded any apparent N-nitrosamines, recovery determinations were completed using 30 replicates from this treatment group.

6. Clostridium botulinum Test

Initially, the effectiveness of the various nitrite-sorbate combinations in the inhibition of C. botulinum growth and toxinogenesis was evaluated using the same procedures as described for the turkey bologna. However, the inoculated turkey ham slices lost vacuum within five days at 27°C and exhibited rapid microbial degradation, so an alternate method was studied. All the procedures were the same as described for the turkey bologna except that the ham was comminuted, instead of being sliced. Ground ham (100 g) was placed in Vac 4 ® bags (Koch, Kansas City, MO.) and inoculated with a C. botulinum spore suspension which provided a concentration of 100 spores/g of product. Samples were stored and analyzed as described for the turkey bologna.

C. RESULTS AND DISCUSSION

1. Test 1: Determination of Minimum Nitrite Level

a. Organoleptic analysis: Turkey ham chubs manufactured with 0, 20, 40, 60, 100 and 156 mg/kg of sodium nitrite were evaluated at regular intervals by sensory panels for the color, flavor, odor, off-flavor, off-odor and overall acceptability. The influence of various nitrite levels on the mean sensory scores for turkey ham color is shown in Figure 14. Analysis of variance (Appendix Table A-33) indicates that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-34) reveals that turkey ham cured with as little as 20 mg/kg of nitrite was not significantly different ($p < 0.01$) from the reference (156 mg/kg of nitrite). This confirmed the report by DuBose et al. (1981) that cooked, smoked hams prepared with 25, 75 and 156 mg/kg of nitrite were equally acceptable in color. The fact that nitrite-free hams have consistently received lower color ratings than their nitrite treated counterparts has also been reported (Kemp et al., 1974; Eakes and Blumer, 1975a,b; Kemp et al., 1975; DuBose et al., 1981). Despite some influence due to product variability and the inexperience of the panelists, the sensory scores for turkey hams showed that increased nitrite input provided increased color acceptability. Similar results have been reported by Brown et al. (1974), Eakes and Blumer (1975a,b) and MacDonald (1978).

The influence of various nitrite levels on the mean panel scores for turkey ham flavor and aroma is shown in Figures 15 and 16, respectively. The analyses of variance (Appendix Tables A-35 and A-36) reveals that only sample treatment (B) was significant ($p < 0.01$) for both

Figure 14: Effect of various nitrite levels on the mean sensory scores for turkey ham color during storage at 4°C.

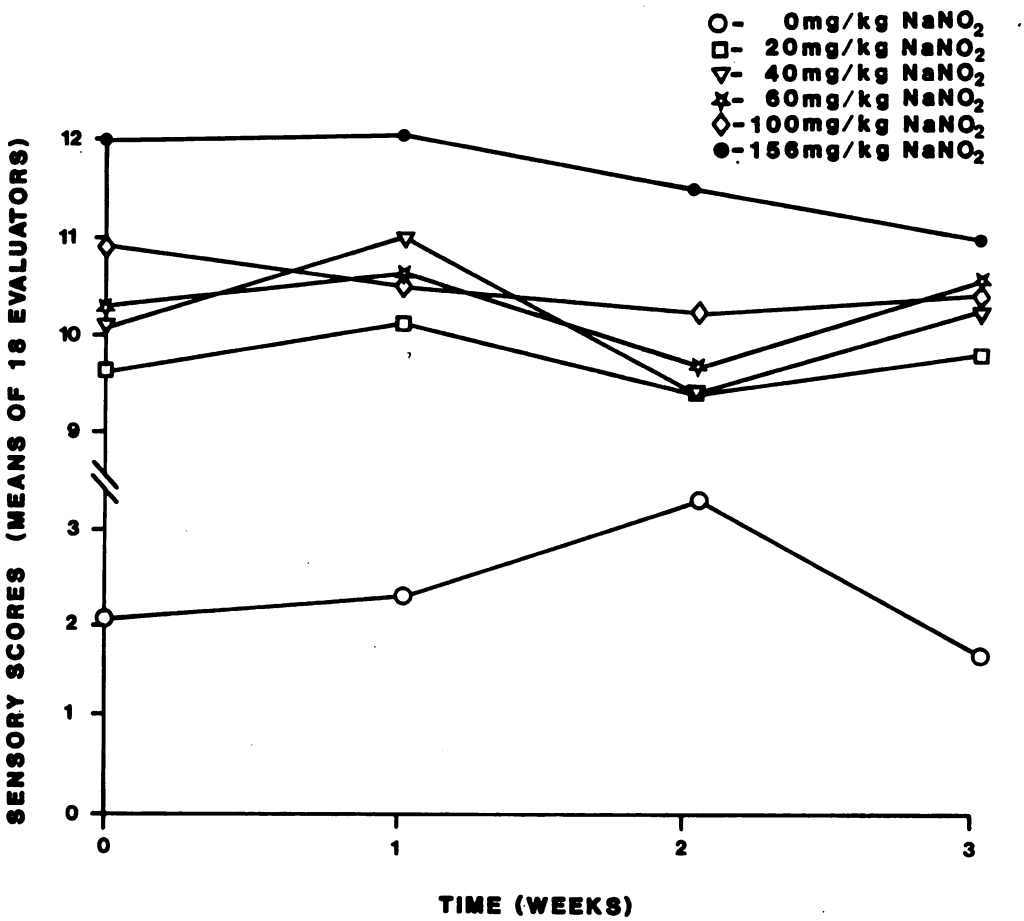

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 18 EVALUATORS)
12
11
10
9
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 14

Figure 15: Effect of various nitrite levels on the mean sensory scores for turkey ham flavor during storage at 4°C.

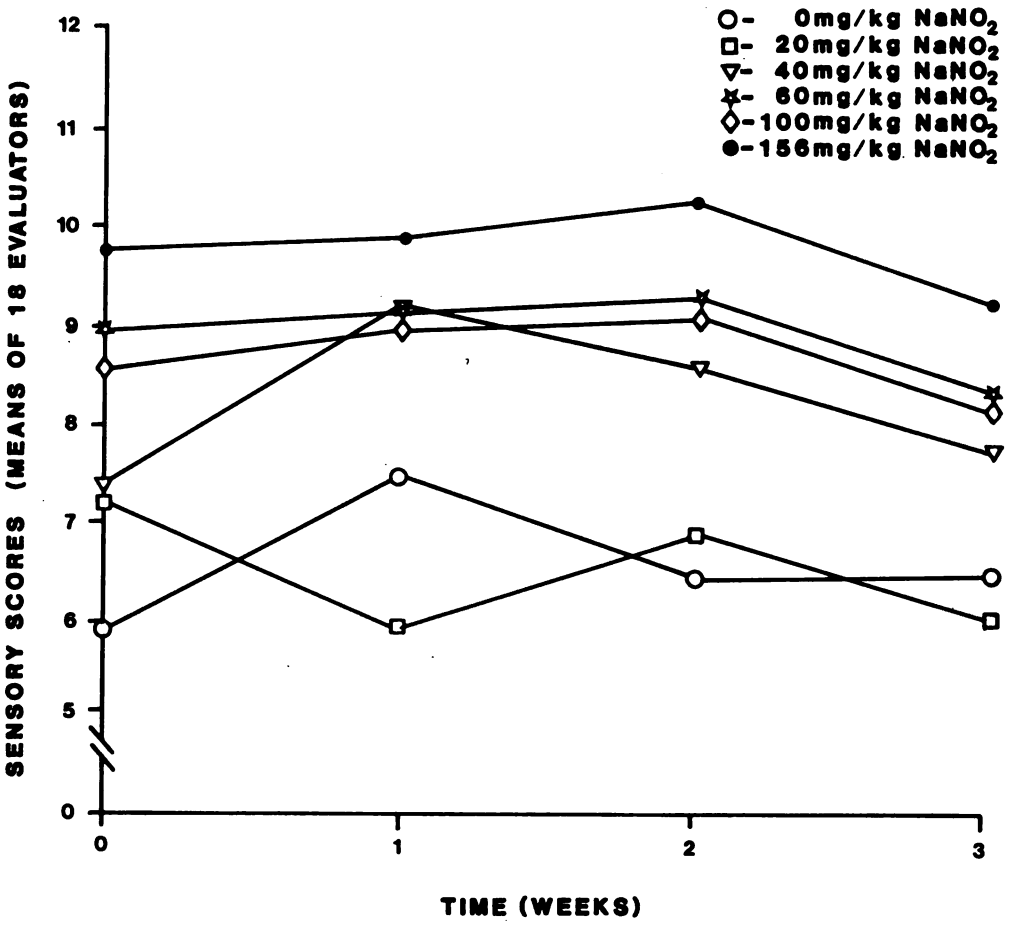

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 18 EVALUATORS)
12
11
10
9
8
7
6
5
0
0
1
2
3
TIME (WEEKS)

Figure 15

Figure 16: Effect of various nitrite levels on the mean sensory scores for turkey ham odor during storage at 4°C.

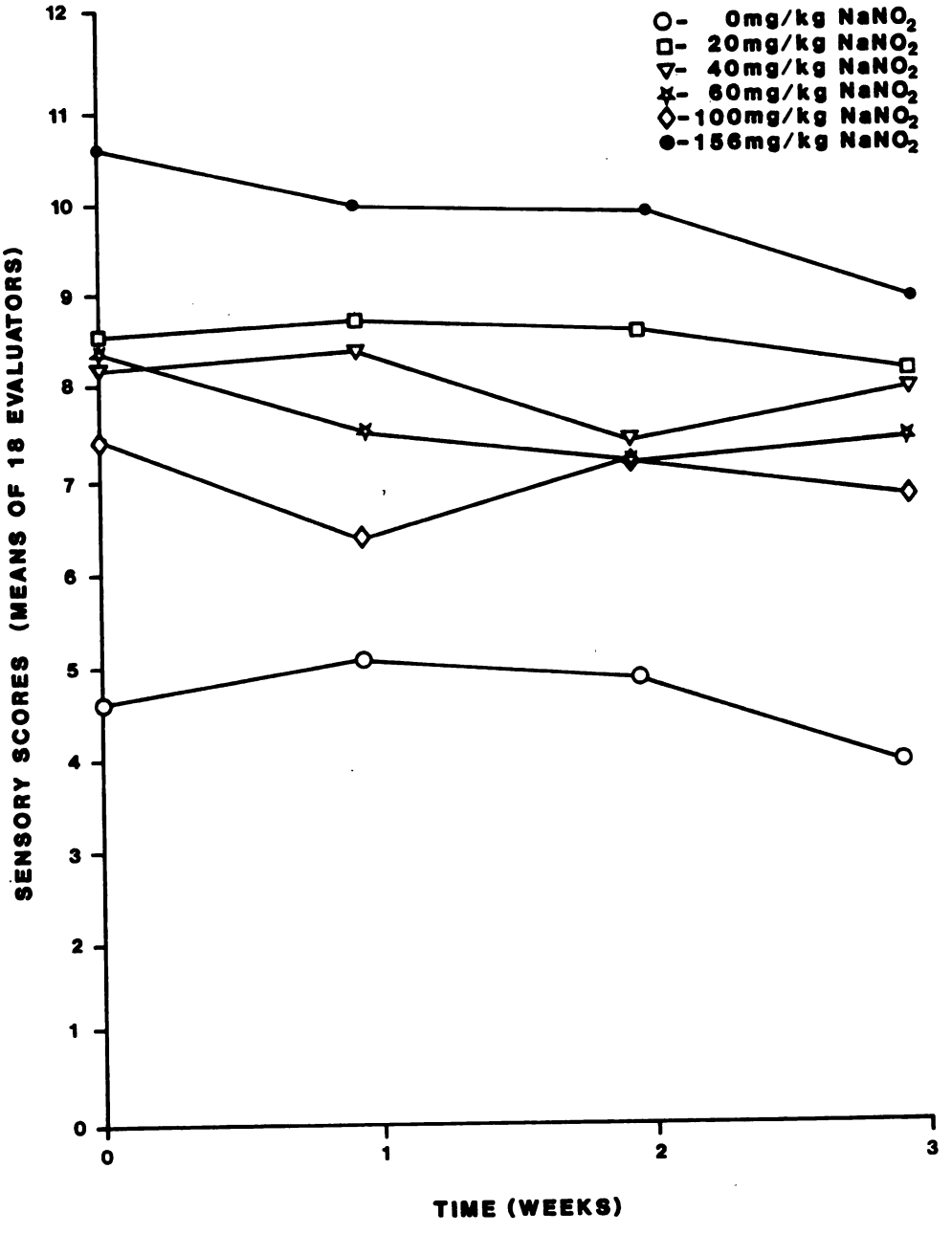

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 18 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
6
7
8
9
10
11
12

Figure 16

attributes. Comparison of treatment means for the flavor scores (Appendix Table A-37) indicates that at least 40 mg/kg of nitrite was necessary to provide a product that was comparable to the reference (156 mg/kg of nitrite). However, for the odor scores, 20 mg/kg of nitrite provided a significantly ($p < 0.01$) acceptable ham (Appendix Table A-38). These results were reflected in the comments from panelists that nonnitrite treated samples exhibited a strong "turkey" or "poultry" flavor, along with a fresh meat odor. The product prepared with 20 mg/kg of nitrite had discernible cured ham characteristics, but the "turkey" flavor was still detectable and the product developed a "tinny", stale aftertaste during refrigerated storage.

The fact that hams cured with nitrite exhibited more intense cured meat flavor than hams cured without nitrite has been reported by many other investigators (Bailey and Swain, 1973; Brown et al., 1974; Eakes and Blumer, 1975a,b; Kemp et al., 1975). MacDonald et al. (1980c) observed that increased levels of nitrite to a concentration of 500 mg/kg in stitch-pumped hams significantly ($p < 0.05$) produced a more intense cured aroma and flavor. However, they also found that nitrite levels as low as 50 mg/kg were sufficient to provide a significant ($p < 0.05$) cured meat flavor when compared to samples containing only salt. Similarly, DuBose et al. (1981) reported that hams cured with 25, 75 and 156 mg/kg of nitrite were equally acceptable in flavor.

Obviously, a ham-type product with organoleptically acceptable cured flavor and aroma can be produced with nitrite levels lower than the current legal limit (156 mg/kg). However, it was interesting to note that in this turkey ham study less nitrite (20 mg/kg) was necessary to

produce an acceptable ham aroma, than was required to provide significant ($p < 0.01$) cured flavor. Possibly the co-presence of a slight "poultry" flavor in the samples containing 20 mg/kg of nitrite impaired the panelist's judgment and thereby increased the level of nitrite necessary for flavor differentiation between treatment groups (Price and Greene, 1978). Another explanation could be that nitrite does not influence cured ham aroma as much as it effects the flavor attribute. In their study of dry-cured hams, Kemp et al. (1974) reported that an excellent ham aroma was found in all treatment groups (with and without nitrite), but that nitrite and/or nitrate had to be present for acceptable flavor development.

The influence of various nitrite levels on the mean sensory scores for turkey ham off-flavor and off-odor is shown in Figures 17 and 18, respectively. The analyses of variance (Appendix Tables A-39 and A-40) reveals that only sample treatment (B) had a significant effect ($p < 0.01$) on either attribute. Comparison of treatment means over time for off-flavor and off-odor scores (Appendix Tables A-41 and A-42, respectively) indicates that only the nonnitrite treated turkey ham was significantly different ($p < 0.01$) from the reference (156 mg/kg of nitrite) for both attributes. Panelists commented that the ham prepared without sodium nitrite exhibited a stale, oxidized aftertaste which increased with storage time. In addition, the "turkey" or "poultry" flavor detected in the product probably contributed to or was interpreted as an undesirable characteristic.

The dramatic reduction in off-flavor and off-odor scores when as little as 20 mg/kg of nitrite was added to the turkey ham samples

Figure 17: Effect of various nitrite levels on the mean sensory scores for turkey ham off-flavor during storage at 4°C.

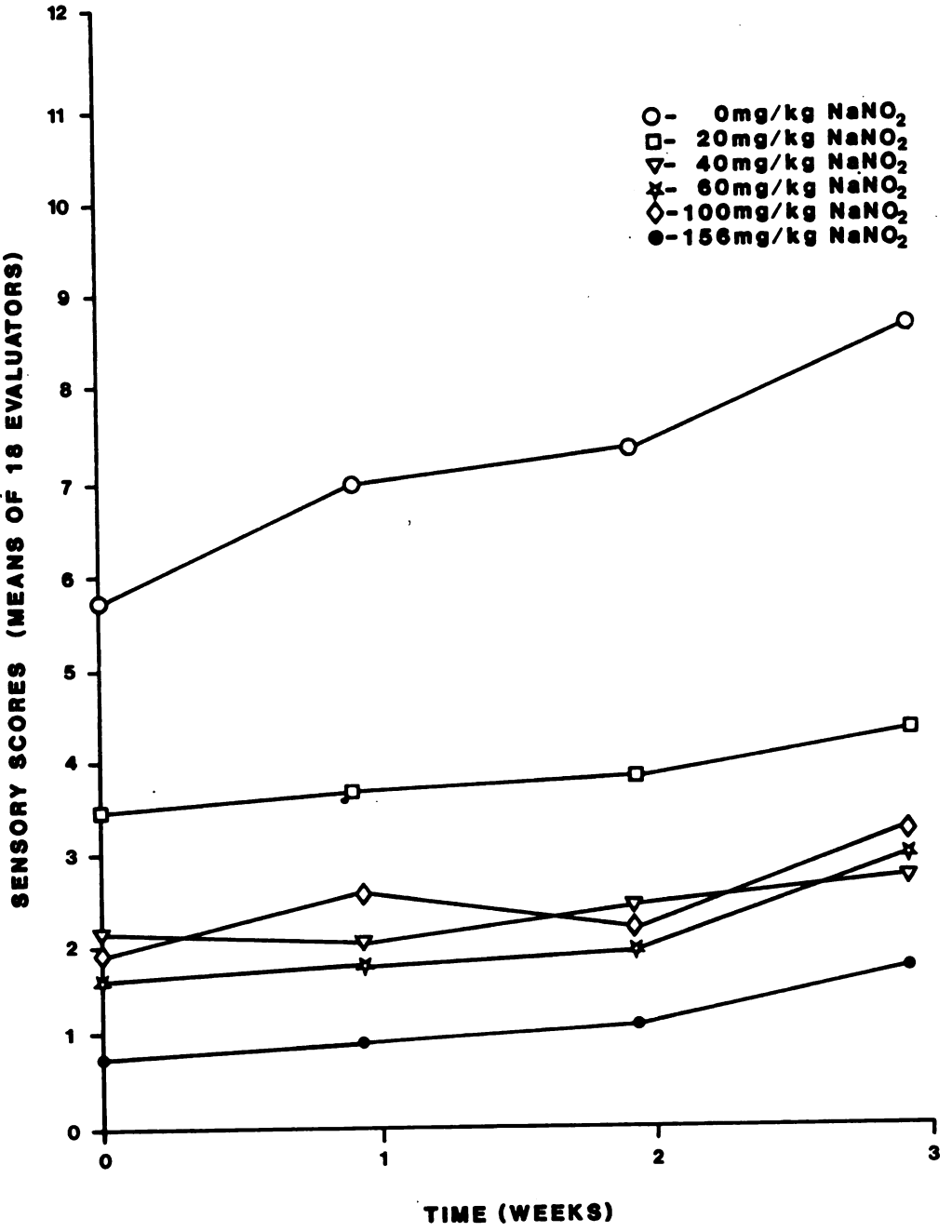

12
11
10
9
8
7
6
5
4
3
2
1
0
SENSORY SCORES (MEANS OF 18 EVALUATORS)
0
1
2
3
TIME (WEEKS)
O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2

Figure 17

Figure 18: Effect of various nitrite levels on the mean panel scores for turkey ham off-odor during the storage at 4°C.

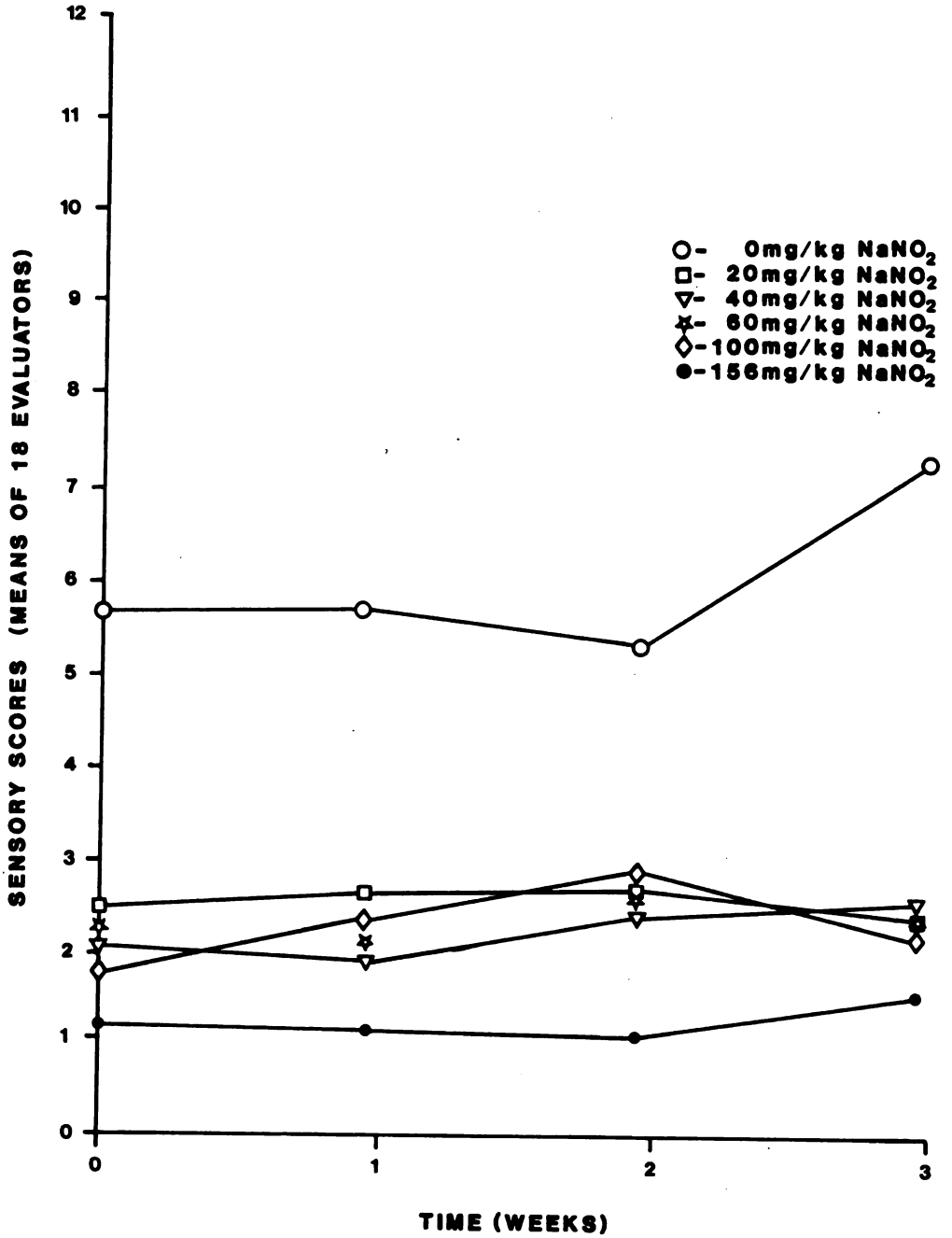

SENSORY SCORES (MEANS OF 18 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
6
7
8
9
10
11
12
O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2

Figure 18

effectively demonstrates the antioxidant activity of nitrite. The ability of nitrite to significantly reduce the development of odors and flavors related to lipid oxidation and/or warmed-over flavor has been shown by several researchers (Zipser et al., 1964; Olson et al., 1978; Price and Greene, 1978; MacDonald et al., 1980c). It is interesting to note that Eakes and Blumer (1975a) reported that the presence or absence of nitrate and/or nitrite in country-style hams did not affect the occurrence of off-flavors as evaluated by panel members. One must consider though, that an oxidized, rancid flavor is a desirable characteristic in aged, country-style hams.

The influence of various nitrite levels on the mean panel scores for the overall acceptability of turkey hams is shown in Figure 19. Analysis of variance (Appendix Table A-43) indicates that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-44) reveals that turkey ham cured with 40 mg/kg or more of nitrite were not significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite). Although the ham prepared with 20 mg/kg of nitrite exhibited much greater acceptability than the nonnitrite treated product, it was still significantly ($p < 0.01$) different from the reference. These results are similar to those reported by Kemp et al. (1975), MacDonald et al. (1980c) and DuBose et al. (1981).

Of the six attributes evaluated by the panel, color probably exerted the greatest influence on the acceptance of a product. This was reflected by remarks from the panelists that the nonnitrite treated turkey ham would have received higher ratings for its other characteristics had

Figure 19: Effect of various nitrite levels on the mean sensory scores for the overall acceptability of turkey hams during storage at 4°C.

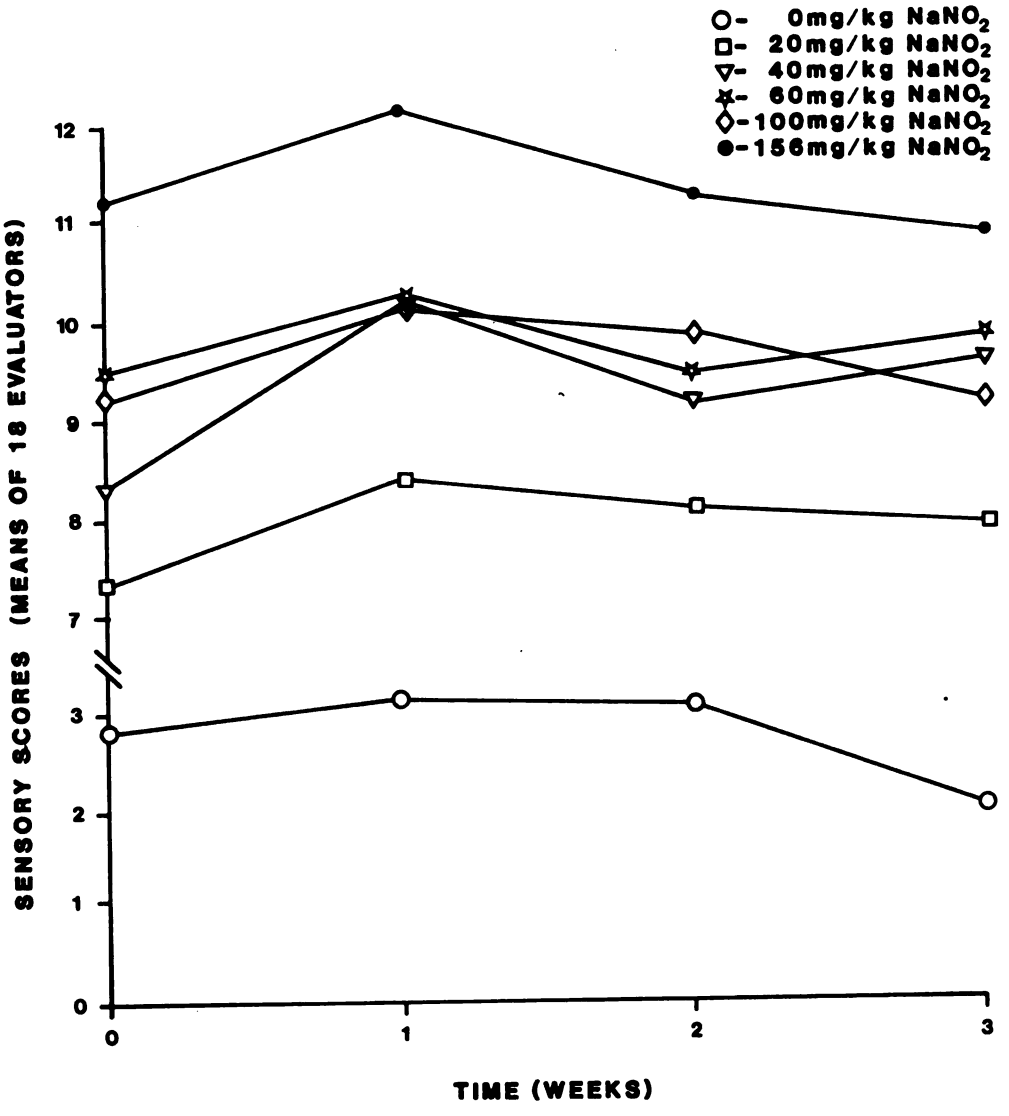

O- 0mg/kg NaNO2
□- 20mg/kg NaNO2
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
◇-100mg/kg NaNO2
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 18 EVALUATORS)
12
11
10
9
8
7
3
2
1
0
0
1
2
3
TIME (WEEKS)

Figure 19

the color been more aesthetically pleasing. Similarly, DuBose et al. (1981) reported that satisfactory color in a product can significantly increase its acceptability, even when other attributes (e.g., flavor) of the product are inferior.

In general, as increased levels of nitrite were added to the turkey ham, consistent trends were observed in the scores for all sensory attributes. However, the ham processed with 100 mg/kg of nitrite was the exception as it consistently received lower sensory ratings than even the samples prepared with 40 or 60 mg/kg of nitrite. Product variability is probably the explanation because several panelists commented that the ham prepared with 100 mg/kg of nitrite exhibited a "different" texture and was less salty than the other treatment samples.

Linear correlation and linear regression analyses were conducted between the mean sensory scores for various attributes of turkey ham. Table 23 indicates that increasing ham flavor and aroma were correlated with decreasing off-flavor and off-odor. In addition, greater product

Table 23: Linear correlation and linear regression of various sensory attributes of turkey hams containing various levels of sodium nitrite.

| Contrast | | Regression | Correlation |
|---|---|---|---|
| Flavor | VS Acceptability | Y = -4.81 + 1.64 X | + 0.77 ** |
| Color | VS Acceptability | Y = 0.60 + 0.85 X | + 0.97 *** |
| Odor | VS Acceptability | Y = -1.53 + 1.31 X | + 0.80 *** |
| Off-Flavor | VS Acceptability | Y = 12.47 - 1.31 X | - 0.95 *** |
| Off-Odor | VS Acceptability | Y = 13.11 - 1.71 X | - 0.94 *** |
| Off-Flavor | VS Off-Odor | Y = 0.52 + 0.72 X | + 0.95 *** |
| Off-Flavor | VS Flavor | Y = 12.81 - 1.21 X | - 0.78 ** |
| Off-Flavor | VS Odor | Y = 10.66 - 1.00 X | - 0.83 *** |
| Off-Odor | VS Odor | Y = 8.93 - 0.82 X | - 0.89 *** |
| Off-Odor | VS Flavor | Y = 8.92 - 0.77 X | - 0.65 ** |
| Odor | VS Flavor | Y = 4.87 + 0.42 X | + 0.54 ** |

***($p < 0.001$)
**($p < 0.01$)

acceptability was associated with increased cured flavor/odor intensity or with decreased off-flavor/off-odor intensity. This confirms the earlier reports by Kemp et al. (1974), Price and Greene (1978) and MacDonald et al. (1980c). The correlations involving the flavor attribute were less significant ($p < 0.01$) than the rest because the flavor results did not exhibit a large differential between the scores for the ham prepared without nitrite and the other nitrite treatments. A vastly larger difference was evident in the analyses of the other sensory attributes.

b. Proximate analysis and pH: Table 24 lists the proximate analyses for the raw turkey thigh meat and the nonnitrite treated turkey ham product. These results are similar to values reported for turkey thigh meat (Uebersax et al., 1978) and for turkey ham (Acton et al., 1979; Bowers et al., 1979). It is interesting to note the differences between various types of meat and processing procedures. Stitch-pumped hams manufactured from pork usually have a lower moisture content (66-68%) than turkey hams (Eakes and Blumer, 1975a), and dry-cured pork hams are even drier (60-65%) (Kemp et al., 1974; Eakes and Blumer, 1975a,b).

Table 24: Proximate analysis of turkey thigh meat and nonnitrite treated turkey ham.

| Sample | Moisture (%) | Fat (%) | Protein (%) |
|---|---|---|---|
| Raw meat | 75.70 ± 0.37 | 5.50 ± 0.38 | 20.10 ± 0.08 |
| Turkey ham | 73.60 ± 0.54 | 8.10 ± 1.20 | 17.30 ± 0.65 |

The pH readings for turkey ham chubs prepared with various levels of nitrite and stored for three weeks at 4°C are listed in Table 25.

Table 25: pH values for turkey ham prepared with various levels of nitrite and stored at 4°C over three weeks.

| Nitrite (mg/kg) | Time (weeks) 0 | 1 | 2 | 3 | X |
|---|---|---|---|---|---|
| 00 | 6.25 | 6.20 | 6.30 | 6.25 | 6.25 ± 0.04 |
| 20 | 6.30 | 6.25 | 6.34 | 6.25 | 6.29 ± 0.04 |
| 40 | 6.34 | 6.25 | 6.33 | 6.25 | 6.29 ± 0.05 |
| 60 | 6.33 | 6.25 | 6.34 | 6.25 | 6.29 ± 0.05 |
| 100 | 6.31 | 6.35 | 6.34 | 6.25 | 6.31 ± 0.05 |
| 156 | 6.33 | 6.30 | 6.34 | 6.25 | 6.31 ± 0.04 |

Although the pH was slightly higher for the ham prepared with 20 mg/kg of nitrite compared to the nonnitrite treated product, there was little difference between the pH values of samples processed with various levels of nitrite. Over the three week storage period, the pH readings remained fairly stable, regardless of sample treatment. These results compare favorably to those reported by Kemp et al. (1974) and Eakes and Blumer (1975a,b). Kemp et al. (1974) observed that pH was significantly correlated with cured color ($p < 0.01$), general appearance ($p < 0.01$), flavor ($p < 0.01$) and overall satisfaction ($p < 0.05$). In general, the higher pH hams (6.0 to 6.2) were more desirable.

c. Residual nitrite: Analysis for residual nitrite in turkey ham during a three week storage period is shown in Table 26. Although increased nitrite input resulted in increased residual nitrite, the ham prepared with the highest level of nitrite (156 mg/kg) contained only

Table 26: Residual nitrite analysis of turkey ham prepared with various levels of nitrite and stored at 4°C over a three week period.

| Time (weeks) | Nitrite Treatment (mg/kg) | | | | | |
|---|---|---|---|---|---|---|
| | 00 | 20 | 40 | 60 | 100 | 156 |
| Week 0 | 0* | 3.4 | 7.6 | 14.6 | 23.8 | 29.3 |
| Week 1 | 0 | 3.9 | 8.0 | 14.8 | 18.8 | 23.4 |
| Week 2 | 0 | 3.8 | 7.0 | 13.8 | 16.2 | 22.9 |
| Week 3 | 0 | 3.3 | 8.8 | 14.0 | 19.2 | 25.0 |

*Nitrite concentrations in mg/kg.

23-29 mg/kg of residual nitrite. This is confirmed by other reports that only 25-50% of the initial level of nitrite introduced into a cure formulation can be detected in the final product (Brown et al., 1974; Eakes and Blumer, 1975a,b; MacDonald, 1978). No detectable residual nitrite was found in the turkey ham prepared without added sodium nitrite. Other authors have reported small levels of nitrite (4-10 mg/kg) in nonnitrite treated hams, but they contended that it could be due to experimental error, natural occurrence or due to cross-contamination as all the treatment samples were cured in the same cooler by the same workers (Kemp et al., 1974, 1975). Doerr et al. (1981) observed that the AOAC (Griess) method for residual nitrite analysis could provide low results when both nitrite and a reductant, such as ascorbate, are simultaneously present. They suggested that ascorbate, and to a lesser extent other reductants, compete with the aromatic amines for the nitrosating species ($N_2O_3$).

The residual nitrite levels in the turkey ham samples decreased over storage time, with the largest loss during the first week.

Nordin (1969) observed a similar depletion of sodium nitrite during the curing, cooking and storage of ground ham meat. He reported that as the level of nitrite decreased, the rate of depletion decreased until at low levels almost no change occurred. Furthermore, the rate of nitrite depletion is exponentially related to both temperature and pH and doubles for every 12.2°C (22°F) increase in temperature or 0.86 units decrease in pH (Nordin, 1969).

d. Hunter colorimetry: The color of turkey ham chubs manufactured with various levels of sodium nitrite was objectively analyzed for the L (lightness), $a_L$ (redness), and $b_L$ (yellowness) values. Analysis of variance (Appendix Table A-45) indicates that time (A), treatment (B) and the time-treatment interaction (A X B) were significant ($p < 0.01$) for all three parameters. Comparison of treatment means within each time interval (Appendix Table A-46) reveals that only the nonnitrite treated sample exhibited redness ($a_L$) and yellowness ($b_L$) values which were significantly different ($p < 0.01$) from the reference (156 mg/kg of nitrite). The L values exhibited similar results during the first two weeks of storage, but the mean values were inconclusive by the third week. Increased nitrite input produced increased redness and decreased yellowness in the turkey ham products. Nitrite addition reduced the sample lightness compared to the ham prepared without nitrite (0 mg/kg), but these L values did not exhibit any general trend as the ingoing level of nitrite was increased. Over the three week storage period, the results for all three color parameters remained fairly stable, regardless of sample treatment.

Others have reported a significant ($p < 0.05$) improvement in ham color with increasing levels of nitrite (MacDonald, 1978; Acton et al., 1979). In their study of turkey hams, Acton et al. (1979) observed that a maximum pigment conversion (myoglobin to nitrosylmyoglobin) of 91.3% was attained with 156 mg/kg of nitrite. Furthermore, they established that color development was temperature dependent. From 49°C to 66°C, redness development increased as the internal temperature of the turkey hams increased, and the greatest color formation occurred between 43°C and 49°C.

The significant relationships ($p < 0.001$) between Hunter values and subjective color results are indicated in Table 27. MacDonald (1978) found similar correlations and concluded that Hunter color values are useful indices for color measurement of cured meat systems.

Table 27: Relationship between Hunter values and the subjective evaluations for turkey ham color.

| Contrast | Regression | Correlation |
|---|---|---|
| Color VS Hunter (L) | Y = 43.10 - 0.25 X | - 0.82 *** |
| Color VS Hunter (a) | Y = -6.60 + 1.57 X | + 0.93 *** |
| Color VS Hunter (b) | Y = 35.90 - 3.28 X | - 0.92 *** |

*** ($p < 0.001$)

e. TBA values: Lipid oxidation in turkey ham was objectively analyzed by the TBA method. Analysis of variance (Appendix Table A-47) indicates that time (A), treatment (B) and the time-treatment interaction (A X B) were significant ($p < 0.01$) for this test. Comparison of treatment means within time (Appendix Table A-48) reveals that the introduction

of 20 mg/kg of nitrite dramatically reduced the TBA values compared to the nonnitrate treated sample. However, 40 mg/kg of nitrite was necessary to provide results which were significantly similar ($p < 0.01$) to the reference over the entire three week storage period. As the ingoing level of nitrite was increased, the TBA values subsequently decreased, but this effect was not significant. Many other investigators have observed a reduction in lipid oxidation when nitrite was incorporated into the product formulation (Zipser et al., 1964; Swain, 1972; Olson et al., 1979; MacDonald et al., 1980b). Furthermore, Younathan and Watts (1959) have suggested that nitrite and sodium chloride acted synergistically to retard oxidation in cooked meat stored at refrigerator temperatures.

Over the three week storage period, the TBA values for all turkey ham treatment groups were variable and provided no consistent trends. The TBA test analyzes for malonaldehyde, an unstable product of lipid oxidation. This compound reaches peak concentrations at the same time that oxygen uptake in a product is declining (Hadden et al., 1975). Therefore, the variations in TBA values could be attributed to slight differences in the vacuum levels achieved in individual packages of ham.

The significant ($p < 0.001$) correlations between TBA values and the subjective evaluations for odor, flavor, off-odor, off-flavor and overall acceptability are indicated in Table 28. This indicates that the undesirable organoleptic characteristics of the turkey ham were due to lipid oxidation rather than due to bacterial souring and/or proteolysis. Similar findings were reported by Zipser et al. (1964) and MacDonald et al. (1980b). Both groups concluded that TBA values could

be useful indices for predicting the development of off-odors and flavors in cooked meats during storage.

Table 28: Relationship between TBA values and the subjective evaluations for turkey ham flavor and odor.

| Contrast | Regression | Correlation |
|---|---|---|
| TBA VS Off-Flavor | Y = 1.80 + 1.52 X | + 0.93 *** |
| TBA VS Off-Odor | Y = 1.70 + 1.15 X | + 0.97 *** |
| TBA VS Flavor | Y = 8.60 - 0.63 X | - 0.60 ** |
| TBA VS Odor | Y = 8.50 - 1.10 X | - 0.81 *** |
| TBA VS Acceptability | Y =10.10 - 2.07 X | - 0.92 *** |

***($p < 0.001$)
**($p < 0.01$)

f. N-Nitrosamine analysis: Turkey ham chubs prepared with various nitrite levels were analyzed for volatile N-nitrosamines by GC-TEA. The presumptive results listed in Table 29 indicate that only the ham prepared with 156 mg/kg of nitrite contained detectable N-nitrosamines. This sample exhibited only 0.7 µg/kg of N-nitrosomorpholine (NMOR). Similarly, N-nitrosamine surveys conducted by the Special Poultry Research Committee (Bauermann, 1979) and by the Nitrite Safety Council (1980) have not found detectable N-nitrosamines in cured poultry products. However, Gray et al. (1981a) analyzed chicken frankfurters and observed higher levels of NDMA, NPIP, NPYR and NMOR than reported in other studies. They attributed it to the use of older meat in the manufacture of the chicken frankfurters. Proteolysis, decarboxylation and other effects of storage/age upon meat could enhance N-nitrosamine formation.

Table 29: Presumptive N-nitrosamine levels (μg/kg) in turkey ham prepared with various levels of sodium nitrite.

| Nitrite (mg/kg) | NDMA | NDEA | NDBA | NPIP | NPYR | NMOR |
|---|---|---|---|---|---|---|
| 00 | ND* | ND | ND | ND | ND | ND |
| 20 | ND | ND | ND | ND | ND | ND |
| 40 | ND | ND | ND | ND | ND | ND |
| 60 | ND | ND | ND | ND | ND | ND |
| 100 | ND | ND | ND | ND | ND | ND |
| 156 | ND | ND | ND | ND | ND | 0.7 |

*ND, none detected, less than the limit of detection.

The presence of relatively high levels of NMOR (up to 11 μg/kg) observed in chicken frankfurters (Gray et al., 1981a) and in other cured products (Evans-Holland, 1980) has raised the question as to its mode of formation. An investigation, independent of this thesis research, indicated that the precursor was present in some sanitizers used to clean the stainless steel surfaces in meat processing plants, but in levels too low to account for the NMOR concentration found in cured meats. Rather, it was observed that the largest source was through the steam used in the smokehouse during the cooking cycle. Fajen et al. (1979) have reported that utility steam condensate contained 2 μg/kg of NMOR, probably due to the use of morpholine as a corrosion inhibitor in steam process equipment.

Obviously, there are large differences in the NMOR contents of the turkey ham in this study and the chicken frankfurters analyzed by Gray et al. (1981a). During the GC-TEA analysis, the method of sample preparation may have influenced the determined NMOR levels. Analysis of

a 25 g meat sample involved either a frankfurter measuring 5 inches long and 0.75 inch in diameter, or a ham slice which was 6 inches in diameter and 0.125 inch thick. Although the unit weight analyzed was the same for both products, the frankfurter samples involved over five times the surface area, relative to the initial product, as did the ham slices. Since morpholine was deposited on the surface of the poultry products as a result of steam condensation during the cooking cycle, then higher levels of NMOR would be expected in the chicken frankfurters.

The sensory and chemical analyses of turkey ham chubs prepared with various levels of sodium nitrite indicated that the introduction of 40 mg/kg of nitrite resulted in an organoleptically acceptable product that exhibited reduced TBA values, provided Hunter color values that were not significantly different ($p < 0.01$) from the reference and that did not contain detectable volatile N-nitrosamines. Therefore, the levels of 40 and 60 mg/kg of nitrite were selected for use in Test 2, to be incorporated alone or in combination with potassium sorbate.

2. Test 2: Nitrite-Sorbate Combinations

a. Organoleptic analysis: Turkey ham chubs manufactured with various nitrite-sorbate combinations were evaluated by a sensory panel for color, flavor, odor, off-flavor, off-odor and overall acceptability. The influence of various nitrite-sorbate treatments on the mean sensory scores for turkey ham color is shown in Figure 20. Analysis of variance (Appendix Table A-49) indicates that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-50) reveals that nonnitrite treated samples

Figure 20: Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham color during storage at 4°C.

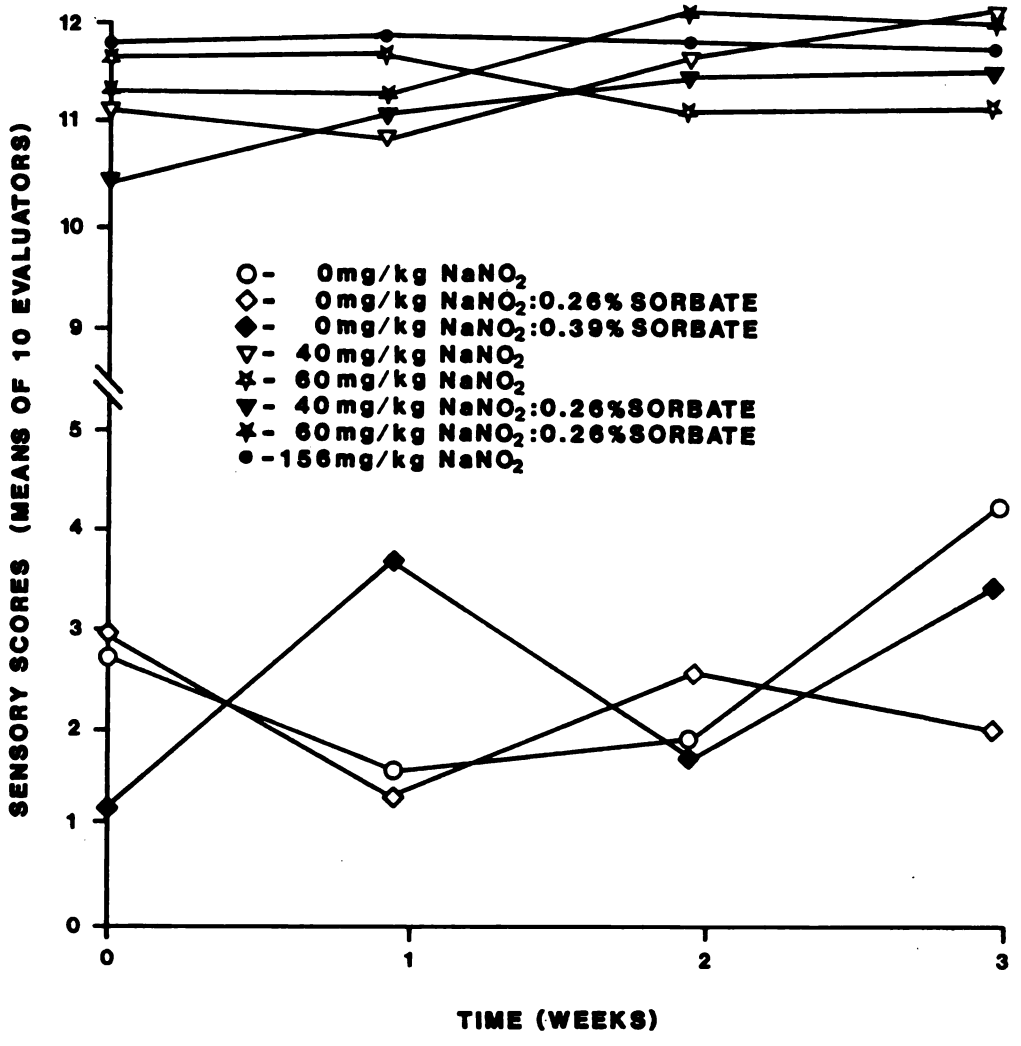

SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
9
10
11
12
○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✲- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✲- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2

Figure 20

and those prepared with sorbate alone (0.26 and 0.39%) exhibited a brownish-grey color and were considered significantly different ($p < 0.01$) from the control (156 mg/kg of nitrite). Panelists commented that the color of turkey ham samples prepared with combinations of 0.26% potassium sorbate and reduced levels of nitrite (40 and 60 mg/kg) was very acceptable. This confirms earlier reports that low levels of nitrite will still provide organoleptically acceptable color (MacDougall et al., 1975; DuBose et al., 1981). Furthermore, these results point out that the addition of sorbate is not a detriment to cured color development; rather, the hams treated with nitrite-sorbate combinations increased in their color acceptability during the three week storage period. Sorbate is known to decrease the rate of nitrite depletion (Sofos et al., 1979c, 1980a), so this affect may have increased the color stability of the nitrite-sorbate treated hams. As the ingoing level of nitrite (alone or combined with 0.26% sorbate) was increased, the subsequent color scores did not exhibit any consistent trends. This serves to emphasize the significant ($p < 0.01$) similarity between the reference and the turkey hams prepared with various nitrite-sorbate combinations.

The influence of various nitrite-sorbate combinations on the mean sensory scores for turkey ham flavor and aroma is shown in Figures 21 and 22, respectively. Analysis of variance (Appendix Tables A-51 and A-52) indicates that only sample treatment (B) was signifcant ($p < 0.01$) for both of these attributes. Comparison of treatment means for flavor and odor scores (Appendix Tables A-53 and A-54, respectively) reveals that the hams prepared without nitrite or with sorbate alone (0.26 and 0.39%) were significantly different ($p < 0.01$) from the reference (156 mg/kg of

Figure 21: Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham flavor during storage at 4°C.

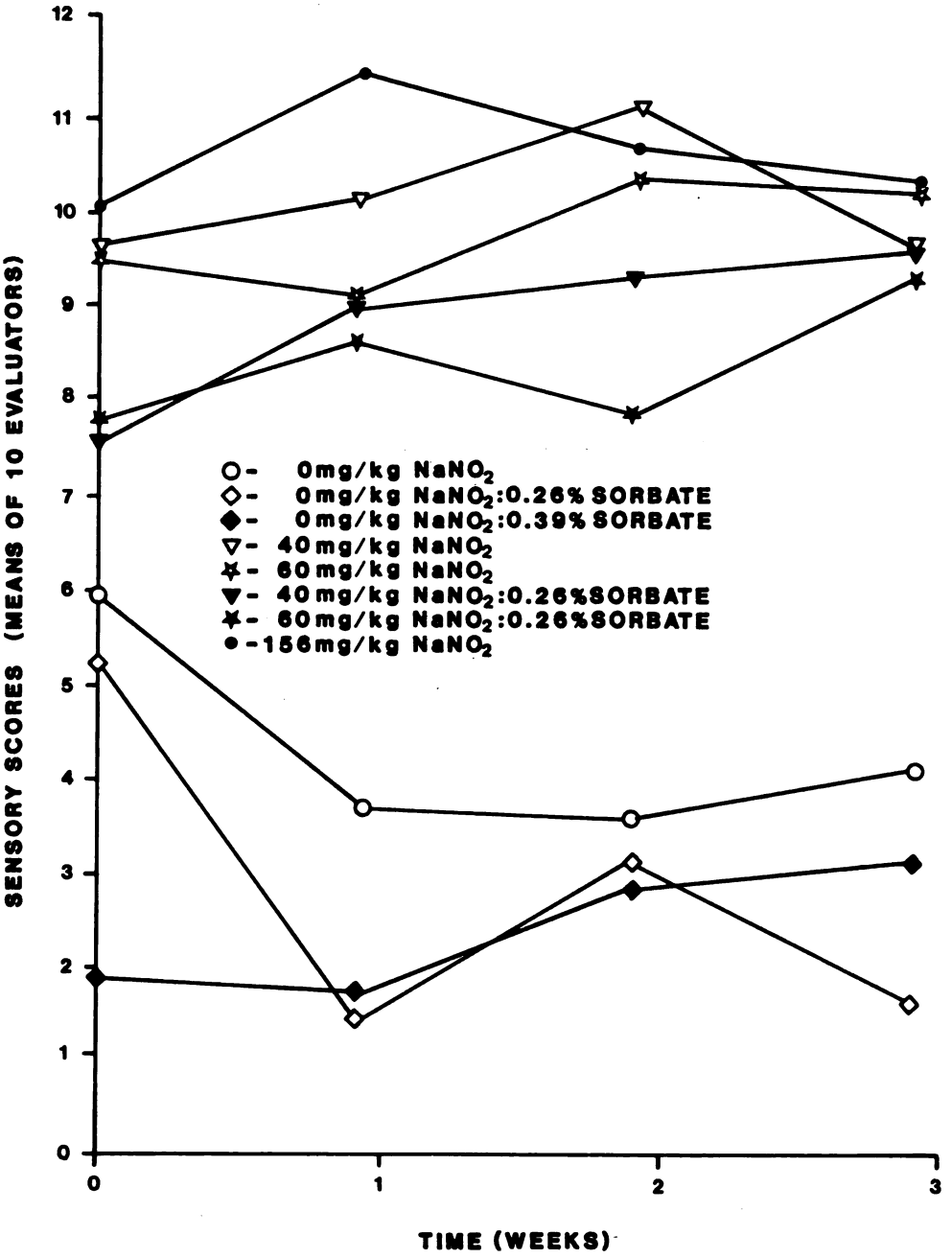

SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
6
7
8
9
10
11
12
○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✱- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2

Figure 21

Figure 22: Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham odor during storage at 4°C.

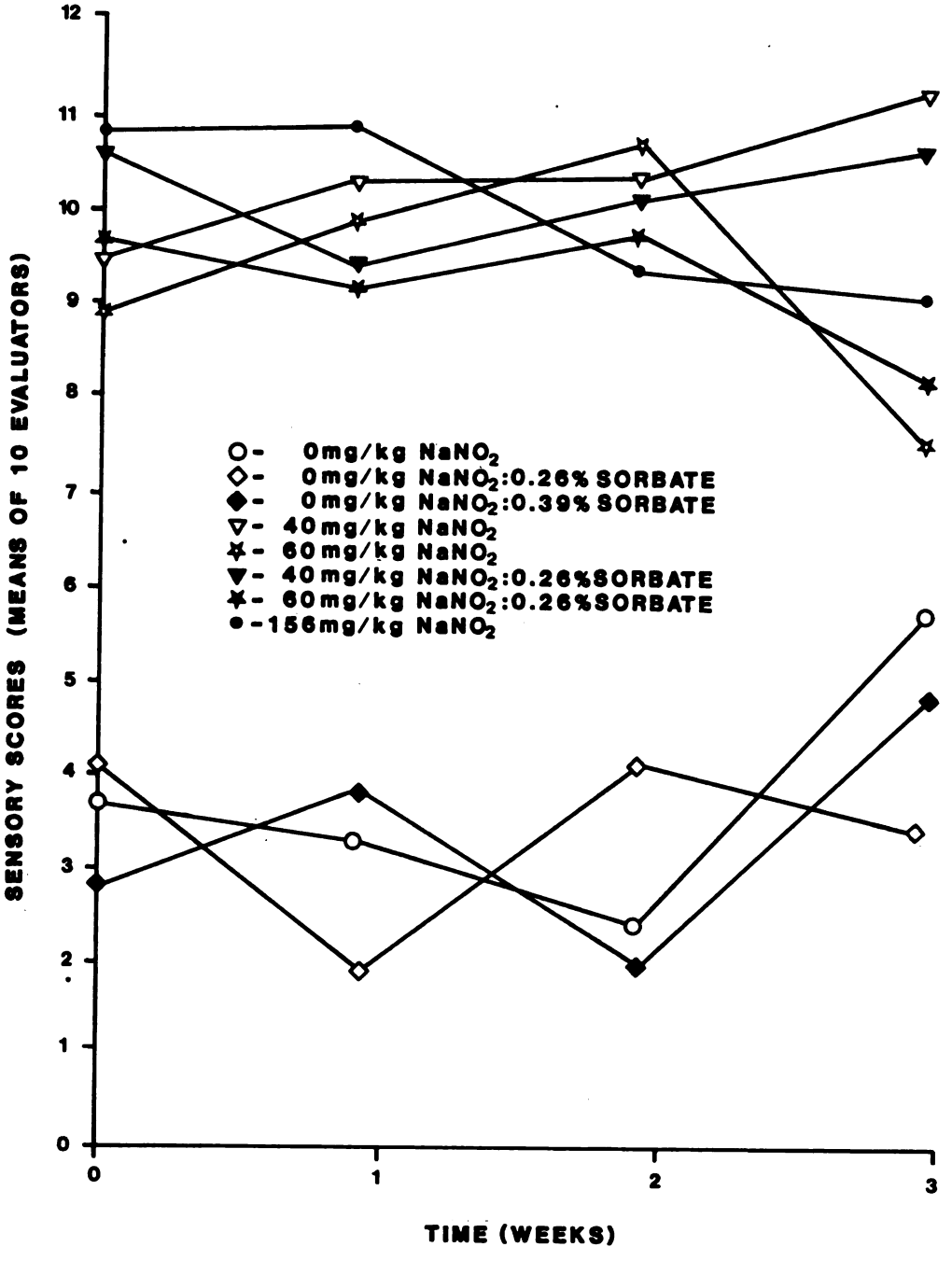

SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
5
6
7
8
9
10
11
12
○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
★- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2

Figure 22

nitrite). Furthermore, the flavor evaluations indicated that the combination of 60 mg/kg of nitrite and 0.26% sorbate was also unacceptable. This treatment combination was similar to the reference when the confidence level was reduced from 99 to 95%, so product variation may have been of influence.

The introduction of sorbate alone into the ham not only failed to provide any detectable cured flavor and aroma, but such samples also received lower flavor scores than hams prepared without nitrite. These results were supported by repetitive comments from the panelists that samples containing sorbate alone exhibited "cardboardy, turkey-like, sweet or tinny" flavors, a "slimy, greasy" mouthfeel and a "strong" odor. When nitrite was combined with sorbate, the "poultry-like" characteristics were undetectable, but a "sweet, saccharin-like" flavor overpowered the cured ham attributes. Overall, sorbate appeared to exert a greater influence on the flavor of the product than its aromatic properties. Panel members commented that sorbate-containing samples that exhibited strong flavors did not necessarily also provide detectable odors.

The influence of various nitrite-sorbate combinations on the mean panel scores for turkey ham off-flavor and off-odor is shown in Figures 23 and 24, respectively. Analysis of variance (Appendix Tables A-55 and A-56) indicates that only the sample treatment (B) was significant ($p < 0.01$) for both of these attributes. Comparison of treatment means for the off-odor evaluations (Appendix Table A-57) reveals that the samples prepared without nitrite (0 mg/kg) or with sorbate alone (0.26 and 0.39%) were significantly ($p < 0.01$) different from the reference (156 mg/kg of nitrite). The same findings were evident for the off-flavor

Figure 23: Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham off-flavor during storage at 4°C.

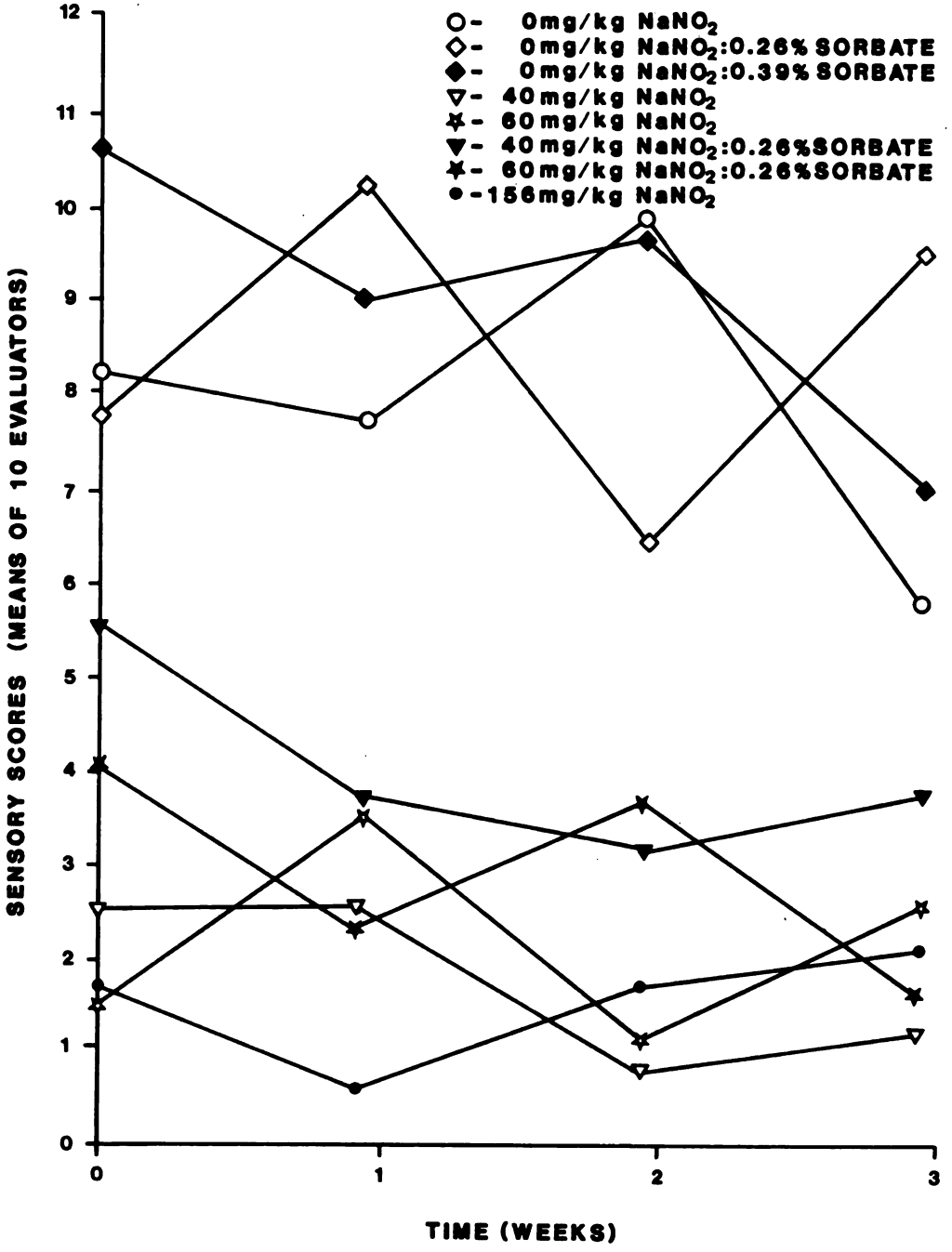

○- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
★- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
0
1
2
3
4
5
6
7
8
9
10
11
12
TIME (WEEKS)

Figure 23

Figure 24: Effect of various nitrite-sorbate treatments on the mean sensory scores for turkey ham off-odor during storage at 4°C.

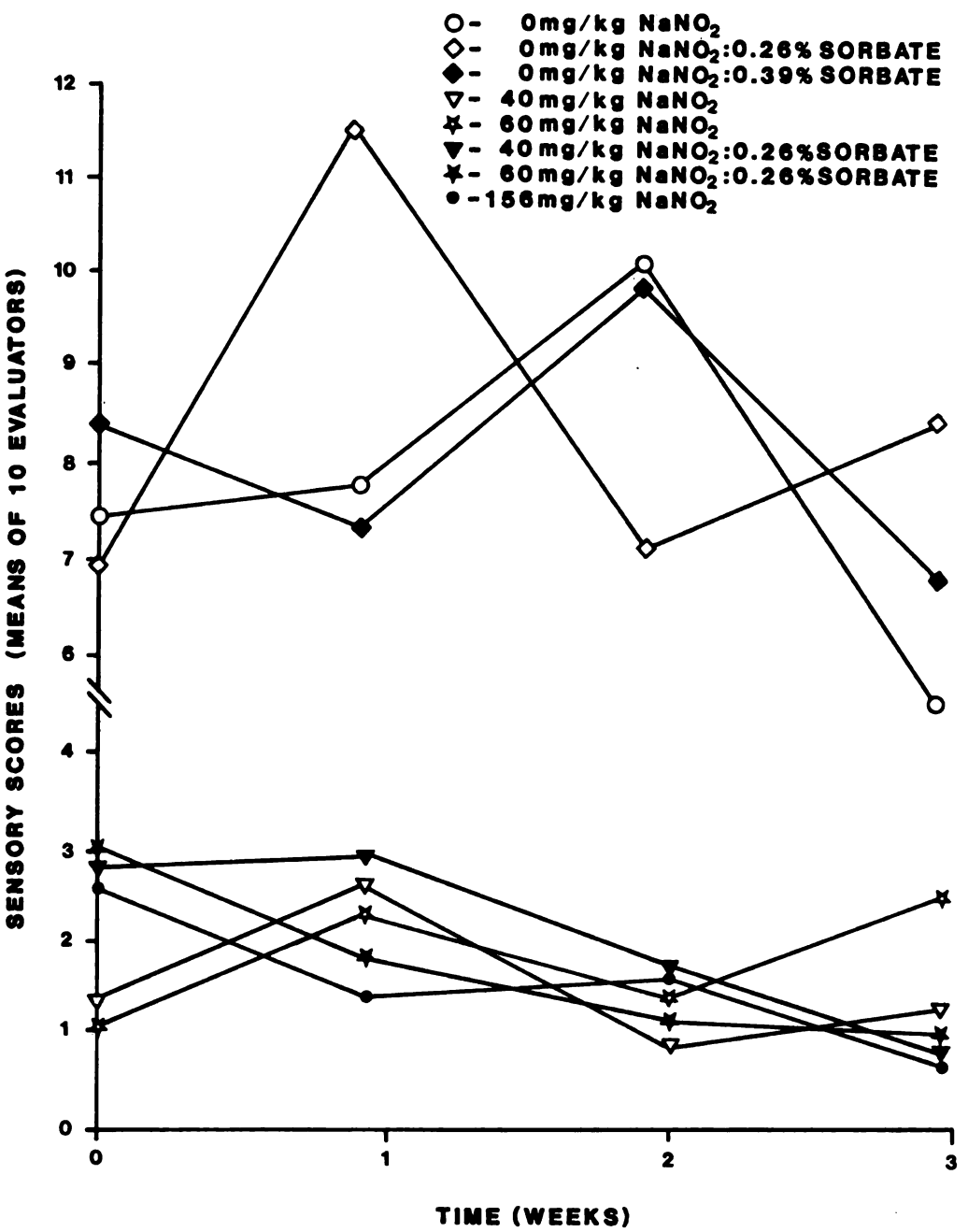

O- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
✲- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✶- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
6
7
8
9
10
11
12

Figure 24

scores (Appendix Table A-58), but for this attribute, the combination 40 mg/kg of nitrite and 0.26% sorbate also resulted in an unacceptable product. Evidently, that level of nitrite was either ineffective against the reaction causing off-flavor production or it could not provide sufficient cured flavor to "mask" the undesirable flavor.

Sorbate itself would appear to be the major contributor to the off-flavor development, since hams prepared with sorbate alone were considered to exhibit greater off-flavor than the samples manufactured without any nitrite. These facts are reflected in the comments from panelists that samples cured with sorbate (either alone or combined with nitrite) had "objectionable, biting" off-flavors and "strong, foul or overpowering" odors.

The influence of various nitrite-sorbate combinations on the mean sensory scores for overall acceptability is shown in Figure 25. Analysis of variance (Appendix Table A-59) indicates that only sample treatment (B) was significant ($p < 0.01$) for this attribute. Comparison of treatment means over time (Appendix Table A-60) reveals that hams cured without nitrite, with sorbate alone (0.26 and 0.39%) or with the combination of 40 mg/kg of nitrite and 0.26% sorbate were all significantly different ($p < 0.01$) from the reference. Similar to the observations for flavor and off-flavor, turkey hams processed with only sorbate received lower acceptability scores than even the samples prepared without any nitrite. Panelists commented that color and flavor were the major determinants of product acceptance. With the exception of the color evaluation, the organoleptic results indicated that turkey ham prepared with a nitrite-sorbate combination was usually rated lower than the

Figure 25: Effect of various nitrite-sorbate treatments on the mean sensory scores for the overall acceptability of turkey ham stored at 4°C.

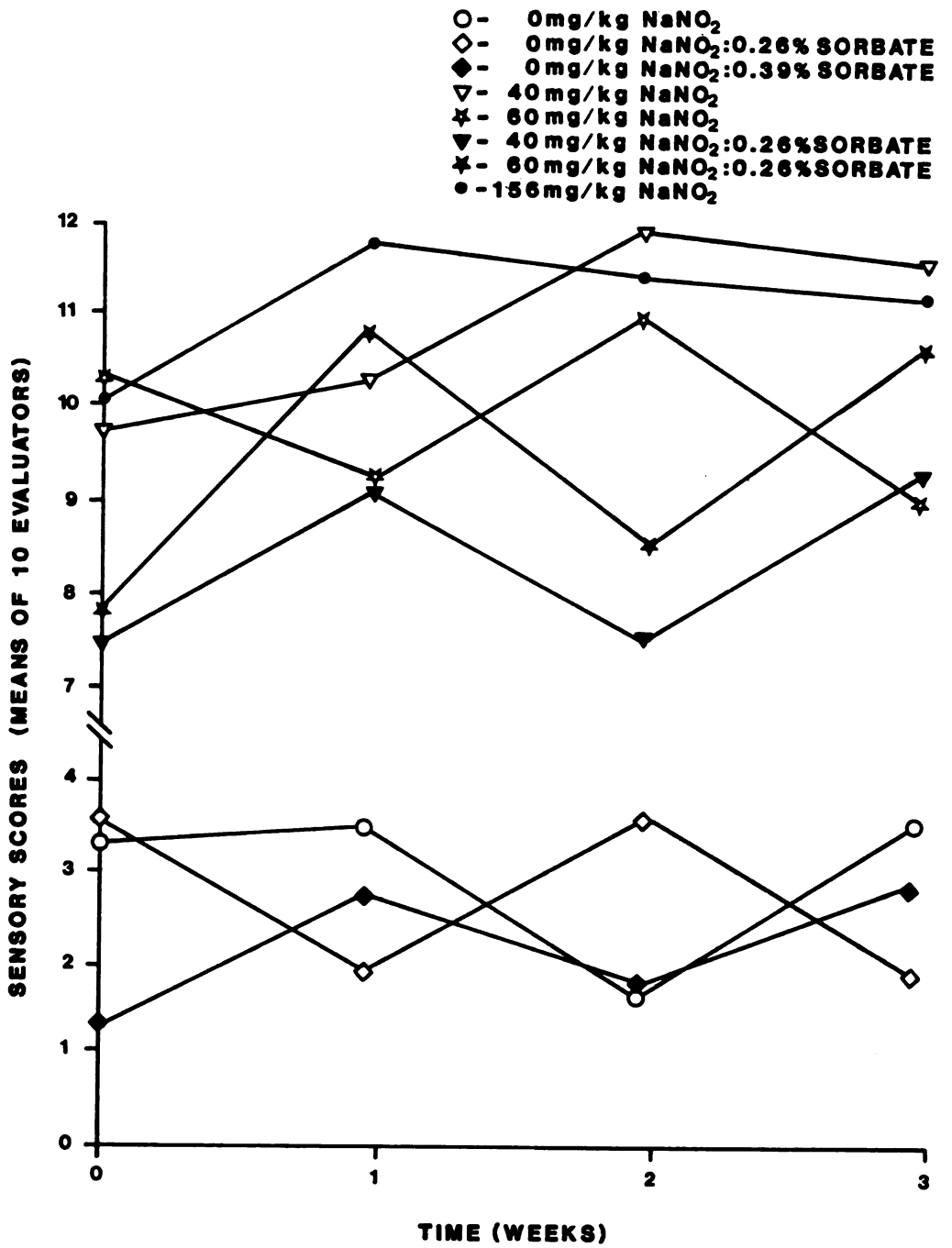

O- 0mg/kg NaNO2
◇- 0mg/kg NaNO2:0.26% SORBATE
◆- 0mg/kg NaNO2:0.39% SORBATE
▽- 40mg/kg NaNO2
☆- 60mg/kg NaNO2
▼- 40mg/kg NaNO2:0.26%SORBATE
✱- 60mg/kg NaNO2:0.26%SORBATE
●-156mg/kg NaNO2
SENSORY SCORES (MEANS OF 10 EVALUATORS)
TIME (WEEKS)
0
1
2
3
4
7
8
9
10
11
12

Figure 25

corresponding product prepared with nitrite alone. Possible product variability was responsible because the ham cured with 60 mg/kg of nitrite (alone or combined with sorbate) consistently received lower flavor and odor scores and higher off-flavor and off-odor ratings than the product cured with 40 mg/kg of nitrite (alone or combined with sorbate). Another possibility is that a compound of undesirable odor and flavor was produced from the interaction of nitrite and sorbate. However, since sorbate itself was blamed for the increased off-flavor and reduced flavor and overall acceptance of turkey ham treated with sorbate alone, it would be more logical that the reduced scores for nitrite-sorbate combinations were merely due to the contributions from sorbate.

In contrast to the findings of this study, Kemp et al. (1979) reported that dry-cured pork hams dipped in a 2.5% potassium sorbate solution prior to storage were considered to be more desirable in flavor ($p < 0.05$) and overall satisfaction ($p < 0.01$) than products prepared with only nitrite. However, such meat products are usually stored aerobically and the yeast and/or mold growth which results during the aging process can contribute undesirable flavors and aromas. The sorbate dip significantly ($p < 0.05$) inhibited such microbial action, so the cured flavor was easier to detect and the sorbate treated hams were considered more acceptable. It also must be considered that only a small portion (<0.5%) of the sorbate in such dipping solutions is actually deposited on the meat surface (Robach et al., 1980a). Therefore, the sorbate levels in the hams studied by Kemp et al. (1979) were probably too low for panelists to detect any off-flavor contributions from sorbate itself.

Linear correlation and linear regression analyses were conducted between mean sensory scores for various attributes. The significant ($p < 0.001$) relationships listed in Table 30 indicate that increasing ham flavor and aroma were associated with decreasing off-flavor and off-odor. In addition, greater product acceptability was correlated with increased flavor/aroma intensity and decreased off-odor/off-flavor intensity.

Table 30: Linear correlation and linear regression between various sensory attributes for turkey hams prepared with nitrite-sorbate combinations.

| Contrast | | Regression | Correlation |
|---|---|---|---|
| Flavor | VS Acceptability | Y = -0.59 + 1.10 X | + 0.96 *** |
| Color | VS Acceptability | Y = 0.63 + 0.81 X | + 0.96 *** |
| Odor | VS Acceptability | Y = 0.98 + 0.86 X | + 0.79 *** |
| Off-Flavor | VS Acceptability | Y = 12.38 - 1.13 X | - 0.97 *** |
| Off-Odor | VS Acceptability | Y = 11.48 - 1.07 X | - 0.94 *** |
| Off-Flavor | VS Off-Odor | Y = -0.45 + 0.97 X | + 0.95 *** |
| Off-Flavor | VS Flavor | Y = 11.60 - 0.97 X | - 0.95 *** |
| Off-Flavor | VS Odor | Y = 11.65 - 0.92 X | - 0.92 *** |
| Off-Odor | VS Odor | Y = 11.25 - 0.93 X | - 0.94 *** |
| Off-Odor | VS Flavor | Y = 10.73 - 0.88 X | - 0.91 *** |
| Odor | VS Flavor | Y = 0.15 + 0.94 X | + 0.91 *** |

*** ($p < 0.001$)

b. Proximate analysis and pH: Proximate analysis of raw turkey meat and of the turkey ham prepared without sodium nitrite are listed in Table 31. Similar results have been reported for turkey thigh pieces (Uebersax et al., 1978) and for turkey ham formulations (Acton et al., 1979). The fact that these findings are comparable to the proximate analyses performed in the Test 1 section demonstrates the precision of the laboratory techniques and formulation procedures used in the study.

Table 31: Proximate analysis of turkey thigh meat and nonnitrite treated turkey ham.

| Sample | Moisture (%) | Fat (%) | Protein (%) |
|---|---|---|---|
| Raw meat | 74.50 ± 0.51 | 5.60 ± 0.21 | 17.80 ± 0.32 |
| Turkey ham | 72.10 ± 0.38 | 8.70 ± 0.54 | 15.40 ± 0.61 |

The pH readings for turkey ham prepared with various nitrite-sorbate treatments and stored over a three week period are listed in Table 32. Although pH values increased when minimum levels of nitrite or sorbate were added, little or no change in the results was observed as the ingoing level of nitrite or sorbate was increased. Hams cured with nitrite-sorbate combinations exhibited pH values similar to products containing either additive alone. Over storage time, pH readings decreased regardless of sample treatment. These results are confirmed by the findings of other studies on potassium sorbate (Ivey et al., 1978; Kemp et al., 1979; Sofos et al., 1980b).

Table 32: pH values for turkey ham prepared with various nitrite-sorbate treatments and stored at 4°C for three weeks.

| Treatment | Time (weeks) | | | | |
|---|---|---|---|---|---|
| | 0 | 1 | 2 | 3 | X |
| 00* | 6.45 | 6.35 | 6.40 | 6.40 | 6.40 ± 0.04 |
| 02 | 6.60 | 6.35 | 6.40 | 6.37 | 6.43 ± 0.12 |
| 03 | 6.60 | 6.40 | 6.50 | 6.40 | 6.48 ± 0.09 |
| 40 | 6.55 | 6.35 | 6.45 | 6.40 | 6.44 1 0.09 |
| 60 | 6.50 | 6.45 | 6.40 | 6.35 | 6.43 ± 0.06 |
| 42 | 6.53 | 6.40 | 6.45 | 6.50 | 6.47 ± 0.06 |
| 62 | 6.53 | 6.45 | 6.55 | 6.40 | 6.48 ± 0.07 |
| 156 | 6.57 | 6.40 | 6.45 | 6.45 | 6.47 ± 0.07 |

*See Table 21 for the nitrite-sorbate combinations corresponding to these identification numbers.

c. Residual nitrite: The residual nitrite analysis of turkey hams processed with various nitrite-sorbate treatments is listed in Table 33.

Table 33: Residual nitrite analysis of turkey ham cured with various nitrite-sorbate combinations and stored at 4°C over a three week period.

| Time (weeks) | Nitrite-Sorbate Treatment | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00[a] | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| Week 0 | 0.0[b] | 0.0 | 0.0 | 8.0 | 16.8 | 7.4 | 17.8 | 35.6 |
| Week 1 | 0.0 | 0.0 | 0.0 | 5.3 | 9.0 | 8.2 | 11.4 | 24.8 |
| Week 2 | 0.0 | 0.0 | 0.0 | 10.0 | 10.0 | 6.6 | 8.0 | 24.4 |
| Week 3 | 0.0 | 0.0 | 0.0 | 6.4 | 9.8 | 6.8 | 8.8 | 24.8 |

[a]See Table 21 for the nitrite-sorbate treatments corresponding to these identification numbers.

[b]Nitrite concentrations in mg/kg.

Although increased nitrite input resulted in greater levels of residual nitrite, only 25 to 35 mg/kg of nitrite was detected in ham prepared with the highest level of nitrite (156 mg/kg). The levels of residual nitrite decreased over time, regardless of sample treatment. Similar results have been reported by Ivey et al. (1978) and Sofos et al. (1980b). Sofos et al. (1979c, 1980a) observed that sorbate can delay/retard residual nitrite depletion. In a similar manner, turkey hams cured with nitrite-sorbate combinations exhibited slightly higher residual nitrite concentrations than the corresponding samples prepared with nitrite alone. This difference was not apparent after two weeks of storage, probably due to the gradual loss of sorbate itself.

d. Hunter colorimetry: Color of turkey ham chubs cured with various nitrite-sorbate combinations was objectively analyzed for the L (lightness), $a_L$ (redness), and $b_L$ (yellowness) values. Analysis of variance (Appendix Table A-61) indicates that time (A), treatment (B) and time-treatment interaction (A X B) were significant ($p < 0.01$) for all three parameters. Comparison of treatment means within each time interval (Appendix Table A-62) reveals that turkey hams prepared without nitrite (0 mg/kg) or with sorbate alone (0.26 and 0.39%) were significantly ($p < 0.01$) lighter, more yellow and less red in color than the reference (156 mg/kg of nitrite). A slight increase in redness, along with a simultaneous decrease in yellowness, were observed when the nitrite input increased from 40 to 60 mg/kg. However, this effect was not significant ($p < 0.01$) and little difference in color values was noted between samples prepared with 60 and 156 mg/kg of nitrite. The combination of nitrite and sorbate resulted in greater $a_L$ and lower $b_L$ values than for the corresponding product containing nitrite alone. This result correlates with the greater residual nitrite reported in nitrite-sorbate treated turkey hams. Over the three week storage period, the values for all three color parameters fluctuated and provided no consistent trends.

The significant relationships ($p < 0.001$) between Hunter values and subjective color results are listed in Table 34. It therefore appears that Hunter color values are useful indices for the color measurement of cured meats.

Table 34: Relationship between Hunter values and the subjective evaluation of turkey ham color.

| Contrast | Regression | Correlation |
|---|---|---|
| Color VS Hunter (L) | Y = 43.06 - 0.25 X | - 0.82 *** |
| Color VS Hunter (a) | Y = 6.07 + 0.42 X | + 0.83 *** |
| Color VS Hunter (b) | Y = 7.01 - 0.51 X | - 0.79 *** |

***($p < 0.001$)

e. TBA values: Lipid oxidation in turkey ham chubs manufactured with various nitrite-sorbate combinations was objectively analyzed over the three week storage period. Analysis of variance (Appendix Table A-63) indicates that time (A), treatment (B) and the time-treatment interaction (A X B) were all significant ($p < 0.01$) for the TBA results. Comparison of treatment means within each time interval (Appendix Table A-64) reveals the antioxidant activity of nitrite. Only the ham prepared without nitrite or with sorbate alone (0.26 and 0.39%) exhibited TBA values significantly ($p < 0.01$) different from the control. Furthermore, the similarity in the results for samples containing only sorbate compared to the nonnitrite product demonstrates the ineffectiveness of sorbate against lipid oxidation. Increased nitrite input resulted in lower TBA values and this effect was more evident over storage time. During the first week of storage the combination of 60 mg/kg of nitrite and 0.26% sorbate provided lower TBA values than when 60 mg/kg of nitrite was introduced alone. However, this effect was reversed by the second week, probably due to the gradual loss of sorbate and a subsequent reduction in the efficacy of sorbate to delay nitrite depletion. Results for ham

cured with 40 mg/kg of nitrite were different from the corresponding product containing nitrite and 0.26% sorbate, but no consistent trend was observed. In general, the TBA values increased over the three week storage period for all treatment groups. These results confirmed the report by Gray et al. (1979).

The significant ($p < 0.001$) correlations between TBA values and the subjective evaluations for odor, flavor, off-odor, off-flavor and overall acceptability are listed in Table 35. This indicates that the undesirable organoleptic characteristics of turkey ham were attributable to lipid oxidation rather than due to microbial action. Despite their significance, the numerical values of these correlations are still lower than the results reported in the Test 1 section for turkey ham. As it was speculated in previous discussions, possible sorbate is also contributing to the "oxidized, stale" aftertaste and undesirable aromas detected in the product.

Table 35: Relationship between TBA values and subjective flavor and odor evaluations of turkey hams.

| Contrast | Regression | Correlation |
|---|---|---|
| TBA VS Off-Flavor | Y = 1.88 + 1.69 X | + 0.81 *** |
| TBA VS Off-Odor | Y = 1.09 + 1.81 X | + 0.84 *** |
| TBA VS Flavor | Y = 10.19 - 1.89 X | - 0.88 *** |
| TBA VS Odor | Y = 10.45 - 1.78 X | - 0.89 *** |
| TBA VS Acceptability | Y = 10.62 - 2.13 X | - 0.87 *** |

*** ($p < 0.001$)

f. N-Nitrosamine analysis: Turkey ham chubs prepared with various nitrite-sorbate treatments were analyzed for volatile N-nitrosamines. Only the ham prepared with 156 mg/kg of nitrite and no sorbate contained detectable N-nitrosamines. This treatment sample exhibited only 0.7 µg/kg of NMOR. Although the use of nitrite-sorbate combinations resulted in greater residual nitrite levels than observed in products prepared with nitrite alone, the turkey hams manufactured with both preservatives did not exhibit any detectable N-nitrosamines. These results concur with earlier reports that meats processed with potassium sorbate and reduced levels of sodium nitrite exhibit low or zero levels of volatile N-nitrosamines (Ivey et al., 1978; Robach et al., 1980c).

g. Clostridium botulinum study: The efficacy of the various nitrite-sorbate combinations in inhibiting C. botulinum growth and toxinogenesis was evaluated by subjecting inoculated turkey ham slices to temperature abuse at 27°C. Within five days of storage, all treatment samples had lost vacuum. Although such package swelling is not indicative of toxin formation, the rapid microbial degradation of the products after seven days of incubation prohibited toxin analyses. This experiment was repeated three times, using freshly processed turkey ham for each trial, but the same results occurred in every test. A limited number of studies on turkey ham have been published (Acton et al., 1979; Bowers et al., 1979), but none have dealt with C. botulinum toxin production. Furthermore, such difficulties have not been mentioned in C. botulinum studies on other meat products.

It is possible that the gas production was due to indigenous microorganisms, rather than due to rapid C. botulinum growth. Huhtanen

and Feinberg (1980) held uninoculated chicken frankfurter emulsions at 30°C and observed swelling within two days. Culturing of the emulsion on SPS agar indicated that the swelling was caused by Clostridium perfringens. Several authors have observed that C. perfringens is one of the most predominant microorganisms indigenous to turkey meat (Guthertz et al., 1976; McKinley and Avens, 1981) and that it will survive the cooking processes used for many poultry products (Baran and Stevenson, 1975). Sorbic acid has been reported to be effective in controlling many of the indigenous and/or contaminating bacteria found in poultry meat (Huhtanen and Feinberg, 1980). However, the sorbate concentration and degree of acidification necessary for such inhibition were greater than those used in the turkey ham product. In addition, the high moisture level (72%) in the turkey ham samples may have allowed the indigenous microflora to grow too rapidly for the sorbate to be effective.

It was observed during the three unsuccessful tests that the inoculated ham slices began to weep fluid after 3-5 days at incubation at 27°C. Consequently, the fourth and final attempt at evaluating the botulinal effectiveness of various nitrite-sorbate combinations involved inoculation of ground/comminuted turkey ham, rather than the slices. The toxin assays of these samples have not been completed; however, some observations were made on the appearance of samples over the six week incubation period. Turkey ham manufactured without nitrite exhibited swelling and proteolytic degradation after 10 days of temperature abuse. The product prepared with the greatest level of nitrite (156 mg/kg) did not display any gas production until after 21 days, but by the fourth

week (28 days) of storage the sample was beginning to deteriorate and smell putrid. After six weeks (42 days) at 27°C, inoculated turkey ham cured with 0.26% sorbate and reduced levels of nitrite (40 and 60 mg/kg) still retained their vacuum and did not exhibit any microbial degradation.

# V. SUMMARY AND CONCLUSIONS

The primary objective of this study was to determine whether potassium sorbate could be used as a supplement and/or replacement for sodium nitrite in cured poultry products (specifically turkey bologna and turkey ham). This is of great interest in view of the increasing public and governmental concern over the presence of carcinogenic N-nitrosamines in certain cured meats. However, the need to provide an aesthetically-pleasing cured meat product which is also botulinal safe must be considered in the determination of a suitable nitrite substitute.

The sensory and chemical analyses of turkey bologna chubs cured with various levels of sodium nitrite indicated that the introduction of approximately 40 mg/kg of nitrite provided a product that was organoleptically acceptable, contained no detectable N-nitrosamines, exhibited reduced TBA values and had Hunter color results which were not significantly ($p < 0.01$) different from the reference (156 mg/kg of nitrite). When increasing levels of nitrite were incorporated into the cure formulation, the sensory, TBA and Hunter color values improved, but this was not a significant ($p < 0.01$) effect. Similar findings were observed for turkey hams prepared with various levels of sodium nitrite.

The evaluation of turkey bologna chubs manufactured with various nitrite-sorbate combinations revealed that samples prepared without nitrite or with sorbate alone (0.26 and 0.39%) were not organoleptically acceptable and exhibited Hunter color and TBA values which were

significantly ($p < 0.01$) different from the control (156 mg/kg of nitrite). Evidently, sorbate alone cannot produce the characteristic pink color or desirable cured meat flavor which are provided by nitrite. Both the sensory and the TBA results demonstrate that sorbate is not effective in retarding lipid oxidation, and furthermore sorbate itself appears to contribute an objectionable sweet,"saccharin-like" off-flavor to a meat product. However, the incorporation of 40 mg/kg of nitrate combined with 0.26% sorbate resulted in turkey bologna chubs that contained no detectable N-nitrosamines and which exhibited sensory, TBA and Hunter color values comparable to a product cured with the USDA approved level of nitrite (156 mg/kg). Although the samples processed with nitrite-sorbate combinations usually received lower organoleptic scores than the corresponding products cured with nitrite alone, this effect was not significant ($p < 0.01$). Despite its many disadvantages, sorbate does assist nitrite in insuring the botulinal safety of cured poultry products. In fact, the incorporation of sorbate into a cure formulation reduces the level of nitrite required to delay C. botulinum growth and toxinogenesis. Turkey bologna prepared with 60 mg/kg of nitrite combined with 0.26% potassium sorbate was found to be as effective as 156 mg/kg of nitrite in inhibiting C. cotulinum.

For the most part, the results from the sensory, chemical and microbiological analyses of turkey hams prepared with various nitrite-sorbate treatments were similar to those reported for the turkey bologna. However, the subjective evaluations for the flavor, off-flavor and overall acceptability of the turkey hams indicated that the combination of 40 mg/kg of nitrite and 0.26% sorbate resulted in a product which was

significantly ($p < 0.01$) different from the reference. These results emphasize the difference between a comminuted product, such as turkey bologna, and one which is produced by the protein-binding of large meat pieces (turkey ham). Emulsification of meat increases the accessible surface area and enhances distribution of additives. Therefore, it was not surprizing that the noncomminuted product, turkey ham, required greater levels of nitrite (in combination with sorbate) to provide acceptable sensory attributes. The difference between the ham and bologna also points out the need to evaluate the efficacy of a potential nitrite substitute in a wide variety of meats and meat products because each system can present a unique challenge.

The overall results from this study indicate that potassium sorbate has potential as a nitrite supplement. However, there are still many unanswered questions regarding the use of sorbate. Therefore, future studies should focus on several areas including: (1) determination of the source and/or cause of the "sweet, saccharin-like" flavor found in sorbate treated meats, (2) identification of the products from the interaction of sorbate with nitrite and other meat components; such as, amino acids and sulfhydryl groups, (3) evaluation of the carcinogenicity of such products, (4) determination of the degradation products of sorbate in cured meat, and (5) the influence of moisture level and/or water activity ($A_w$) on the botulinal efficacy of sorbate.

# BIBLIOGRAPHY

# BIBLIOGRAPHY


Acton, J. C., Dick, R. L. and Torrence, A. K. 1979. Turkey ham properties on processing and cured color formation. Poultry Sci. 58:843.

Ashworth, J. and Spencer, R. 1972. The perigo effect in pork. J. Fd. Technol. 7:111.

A.O.A.C. 1975. Official Methods of Analysis 12 ed. Association of Official Analytical Chemists, Washington, D.C.

Anonymous. 1976. Potassium sorbate proposed as answer to nitrite reduction in cured meats. Food Process. June, p. 8.

Anonymous. 1978a. Response for bacon. Submitted by American Meat Institute to Hearing Clerk, USDA, Washington, D.C.

Anonymous. 1978b. USDA issues bacon proposal. Food Engineer. June, p. 39.

Anonymous. 1978c. USDA proposes adding sorbate to bacon; nitrite reduced from 120 to 40 ppm. Food Engineer. August, p. 128.

Bailey, M. E., Frame, R. W. and Naumann, H. D. 1964. Studies of the photoxidation of nitric oxide myoglobin. J. Agr. Food Chem. 12:89.

Bailey, M. E. and Swain, J. W. 1973. Influence of nitrite on meat flavor. Proc. Meat Industry Research Conf., American Meat Institute Foundation, Chicago, p. 29.

Baker, R. C., Darfler, J. and Vadehra, D. V. 1969. Type and level of fat and amount of protein and their effect on the quality of chicken frankfurters. Food Technol. 23(6):808.

Baldock, J. D., Frank, P. R., Graham, P. P. and Ivey, F. J. 1979. Potassium sorbate as a fungistatic agent in country ham processing. J. Food Protect. 42:780.

Baran, W. L. and Stevenson, K. E. 1975. Survival of selected pathogens during processing of a fermented turkey sausage. J. Food Sci. 40:618.

Barnes, J. M. and Magee, P. N. 1954. Some toxic properties of N-dimethylnitrosamine. Brit. J. Ind. Med. 11:167.

Batzer, O. F., Santara, A. T., Tan, M. C., Landmann, W. A. and Schweigert, B. S. 1960. Precursors of beef flavor. J. Agr. Food Chem. 8:498.

Bauermann, J. F. 1979. Processing poultry products with and without sodium nitrite. Food Technol. 33(7):42.

Bender, A. E. and Ballance, P. E. 1961. A preliminary examination of the flavor of meat extract. J. Sci. Fd. Agric. 12:683.

Berry, B. W. and Blumer, T. N. 1981. Sensory, physical and cooking characteristics of bacon processed with varying levels of sodium nitrite and potassium sorbate. J. Food Sci. 46:321.

Bharucha, K. R., Cross, C. K. and Rubin, L. J. 1979. Mechanism of N-nitrosopyrrolidine formation in bacon. J. Agr. Food Chem. 27:63.

Bharucha, K. R., Cross, C. K. and Rubin, L. J. 1980. Long-chain acetals of ascorbic and erythorbic acids as antinitrosamine agents for bacon. J. Agr. Food Chem. 28:1274.

Bills, D. D., Hildrum, K. I., Scanlan, R. A. and Libbey, L. M. 1973. Potential precursors of N-nitrosopyrrolidine in bacon and other fried foods. J. Agr. Food Chem. 21:876.

Binkerd, E. F. and Kolari, O. I. 1975. The history and use of nitrate and nitrite in the curing of meats. Fd. Cosmet. Toxicol. 13:655.

Bowen, V. G., Cerveny, J. G. and Deibel, R. H. 1974. Effect of sodium ascorbate and sodium nitrite on toxin formation of Clostridium botulinum in weiners. Applied Microbiol. 27:605.

Bowers, J. A., Gardse, C. and Craig, J. 1979. Sensory characteristics and thiamin content of frozen-stored and unfrozen cured-cooked pork and turkey ham. Poultry Sci. 58:830.

Brooks, J., Hainer, R. B., Morgan, R. B. and Pace, J. 1940. The function of nitrite and bacteria in the curing of bacon and hams. Food Investigation Special Report No. 49. His Majesty's Stationary Office, London.

Brown, C. L., Hedrick, H. B. and Bailey, M. E. 1974. Characteristics of cured ham as influenced by levels of sodium nitrite and sodium ascorbate. J. Food Sci. 39:977.

Cassens, R. G., Greaser, M. L., Ito, T. and Lee, M. 1979a. Reactions of nitrite in meat. Food Technol. 33(7):46.

Cassens, R. G., Ito, T. and Lee, M. 1979b. Morphology of bacon and its possible role in the formation of N-nitrosamines. J. Food Sci. 44:306.

Cassens, R. G., Sebranek, J. G., Kubberod, G. and Woolford, G. 1974. Where does the nitrite go? Food Prod. Develop. 8:50.

Cerveny, J. G. 1980. Effects of change in the production and marketing of cured meats on the risk of botulism. Food Technol. 34(5):240.

Chang, S. S. and Petersen, R. J. 1977. Symposium: The basis of quality in muscle foods. Recent developments in the flavor of meat. J. Food Sci. 42:298.

Cho, I. C. and Bratzler, L. J. 1970. Effect of sodium nitrite on flavor of cured pork. J. Food Sci. 35:668.

Christiansen, L. N. 1980. Factors influencing botulism inhibition by nitrite. Food Technol. 34(5):237.

Christiansen, L. N., Johnston, R. W., Kautter, D. A., Howard, J. W. and Aunan, W. J. 1973. Effect of nitrite and nitrate on toxin production by Clostridium botulinum and on N-nitrosamine formation in perishable canned comminuted cured meat. Applied Microbiol. 25:357.

Christiansen, L. N., Tompkin, R. B. and Sharparis, A. B. 1978. Fate of Clostridium botulinum in perishable canned cured meat at abuse temperatures. J. Food Protect. 41:354.

Christiansen, L. N., Tompkin, R. B., Shaparis, A. B., Johnston, R. W. and Kautter, D. A. 1975. Effect of sodium nitrite and nitrate on Clostridium botulinum growth and toxin production in summer-style sausage. J. Food Sci. 40:488.

Christiansen, L. N., Tompkin, R. B., Shaparis, A. B., Kueper, T. V., Johnston, R. W., Kautter, D. A. and Kolari, O. J. 1974. Effect of sodium nitrite on toxin production by Clostridium botulinum in bacon. Applied Microbiol. 27:733.

Clydesdale, F. M. and Francis, F. J. 1971. The chemistry of meat color. Food Prod. Develop. 6:81.

Coleman, M. H. 1978. A model system for the formation of N-nitroso-pyrrolidine in grilled or fried bacon. J. Fd. Technol. 13:55.

Collins-Thompson, D. L., Chang, P. C., Davidson, C. M., Larmond, E. and Pivnick, H. 1974. Effects of nitrite and storage temperature on the organoleptic quality and toxinogenesis by Clostridium botulinum in vacuum-packaged side bacon. J. Food Sci. 39:607.

Cross, C. K. and Ziegler, P. 1965. A comparison of volatile fractions from cured and uncured meat. J. Food Sci. 30:610.

Cunningham, F. E. 1979. Shelflife and quality characteristics of poultry parts dipped in potassium sorbate. J. Food Sci. 44:863.

Dethmers, A. E. and Rock, H. 1975. Effect of added sodium nitrite and sodium nitrate on sensory qualities and N-nitrosamine formation in thuringer sausage. J. Food Sci. 40:491.

Deuel, H. J. and Alfin-Slater, R. 1954. Sorbic acid as a fungistatic agent for foods. 1. Harmlessness of sorbic acid as a dietary component. Food Res. 19:1.

D.H.E.W., Food and Drug Administration. 1977. Nitrates and nitrites in poultry products. Federal Register 42(171):44376.

Dhillon, A. S. and Maurer, A. J. 1975. Quality measurements of chicken and turkey summer sausages. Poultry Sci. 54:1263.

DiFate, V. G. 1978. Chemical evaluation of sorbic acid/sodium nitrite reaction products: relationship of chemistry to mutagenicity. Monsanto Ind. Chem. Co., St. Louis, MO.

Dimick, P. S., MacNeil, J. H. and Grunden, L. P. 1972. Poultry product quality. Carbonyl composition and organoleptic evaluation of mechanically deboned poultry meat. J. Food Sci. 37:544.

Doerr, R. C., Fox J. B., Lakritz, L. and Fiddler, W. 1981. Determination of nitrite in cured meats by chemiluminescence detection. Anal. Chem. 53:381.

DuBose, C., Cardell, A. V. and Maller, O. 1981. Factors affecting acceptability of low-nitrite smoked, cured ham. J. Food Sci. 46:461.

Duncan, C. L. and Foster, E. M. 1968. Effect of sodium nitrite, sodium chloride and sodium nitrate on germination and outgrowth of anaerobic spores. Applied Microbiol. 16:406.

Dunnett, C. W. 1955. A multiple comparison procedure for comparing several treatments with a control. J. Amer. Stat. Assoc. 50:1096.

Dunnett, C. W. 1964. New tables for multiple comparisons with a control. Biometrics 20:482.

Dwivedi, B. K. 1975. Meat flavor. CRC. Crit. Rev. in Food Technol. 5:487.

Dymicky, M., Fox, J. B. and Wasserman, A. E. 1975. Color formation in cooked model and meat systems with organic and inorganic compounds. J. Food Sci. 40:306.

Eakes, B. K. and Blumer, T. N. 1975a. Effect of nitrate and nitrite on color and flavor of country-style hams. J. Food Sci. 40:973.

Eakes, B. K. and Blumer, T. N. 1975b. Effect of various levels of potassium nitrate and sodium nitrite on color and flavor of cured loin and country-style hams. J. Food Sci. 40:977.

Emard, L. O. and Vaughn, R. H. 1952. Selectivity of sorbic acid media for catalase negative lactic acid bacteria and Clostridia. J. Bacteriol. 63:487.

Ember, L. R. 1980. Nitrosamines: assessing the relative risk. Chem. Eng. News 58(16):20.

Emodi, A. S. and Lechowich, R. V. 1969. Low-temperature growth of Type E Clostridium botulinum spores: effects of sodium chloride, sodium nitrite and pH. J. Food Sci. 34:78.

Ender, F. and Ceh, L. 1971. Conditions and chemical reaction mechanisms by which nitrosamines may be formed in biological products with reference to their possible occurrence in food products. Z. Lebensm.-Unters.-Forsch. 145:133.

Eriksson, C. E., Olson, P. A. and Svensson, S. G. 1971. Denatured hemeproteins as catalysts in lipid oxidation. J. Amer. Oil Chem. Soc. 48:442.

Evans-Holland, G. C. 1980. Survey of N-nitrosamines in Canadian cured meats other than bacon. Meat Packers Council of Canada. Islington, Ontario.

Fajen, J. M., Carson, G. A., Rounbehler, D. P., Fan, T. Y., Veta, R., Goff, U. E., Wolf, M. H., Edwards, G. S., Fine, D. H., Reinhold, V. and Biemann, K. 1979. N-nitrosamines in rubber and tire industry. Science 205:1262.

Fan, T. Y. and Tannenbaum, S. R. 1973. Natural inhibitors of nitrosation reaction: the concept of available nitrite. J. Food Sci. 38:1067.

Fiddler, W., Pensabene, J. W., Fagen, J. C., Tho, E. J., Piotrowski, E. G. and Wasserman, A. E. 1974. The role of lean and adipose tissue on the formation of N-nitrosopyrrolidine in fried bacon. J. Food Sci. 39:1070.

Fiddler, W., Pensabene, J. W., Kishnir, J. and Piotrowski, E. G. 1973a. Effect of frankfurter cure ingredients on N-nitrosodimethylamine formation in a model system. J. Food Sci. 38:714.

Fiddler, W., Pensabene, J. W., Piotrowski, E. G., Doerr, R. C. and Wasserman, A. E. 1973b. Use of sodium ascorbate or erythorbate to inhibit formation of N-nitrosodimethylamine in frankfurters. J. Food Sci. 38:1084.

Fiddler, W., Pensabene, J. W., Piotrowski, E. G., Phillips, J. G., Keating, J., Mergens, W. J. and Newmark, H. L. 1978. Inhibition of formation of volatile N-nitrosamines in fried bacon by use of cure-soluble $\alpha$-tocopherol. J. Agr. Food Chem. 26:653.

Fiddler, W., Piotrowski, E. G., Pensabene, J. W., Doerr, R. C. and Wasserman, A. E. 1972. Effect of sodium nitrite concentration on N-nitrosodimethylamine formation in frankfurters. J. Food Sci. 38:668.

Fine, D. H., Rufeh, F., Lieb, D. and Rounbehler, D. P. 1975. Description of the thermal energy analyzer for trace determination of volatile and nonvolatile N-nitroso compounds. Anal. Chem. 47:1188.

Fishbein, L. 1979. Overview of some aspects of occurrence, formation and analysis of N-nitrosamines. The Sci. Total Environ. 13:157.

Fooladi, M. H., Pearson, A. W., Coleman, T. H. and Merkel, R. A. 1979. The role of nitrite in preventing development of warmed-over flavor. Food Chem. 4:283.

Foreman, J. K. and Goodhead, K. 1975. The formation and analysis of N-nitrosamines. J. Sci. Fd. Agric. 26:1771.

Fox, J. B. 1966. Chemistry of meat pigments. J. Agr. Food Chem. 14:207.

Fox, J. B. and Ackerman, S. A. 1968. Formation of nitrosylmyoglobin. Mechanism of reaction with various reductants. J. Food Sci. 33:364.

Fox, J. B., Fiddler, R. N. and Wasserman, A. E. 1981. Initial reaction intermediates in the oxidation of ascorbate by nitrous acid. J. Food Protect. 44:28.

Fox, J. B. and Thomson, J. S. 1963. Formation of bovine nitrosylmyoglobin: pH 4.5-6.5. Biochem. 2:465.

Fox, J. B., Townsend, W. E., Ackerman, S. A. and Swift, C. E. 1967. Cured color development during frankfurter processing. Food Technol. 21(3):386.

Froning, G. W., Arnold, R. G., Mandigo, R. W., Neth, C. E. and Hartung, T. E. 1971. Quality and storage stability of frankfurters containing 15% mechanically deboned turkey meat. J. Food Sci. 36:974.

Gaunt, I. F., Butterworth, K. R., Hardy, J. and Gangolli, S. D. 1975. Long-term toxicity of sorbic acid in the rat. Fd. Cosmet. Toxicol. 13:31.

Giddey, C. 1966. The change in meat pigments in sausage making process. J. Sci. Fd. Agric. 17:14.

Giddings, G. C. 1974. Reduction of ferrimyoglobin in meat. CRC Crit. Rev. Fd. Technol. 5:143.

Giddings, G. C. 1977a. The basis of color in muscle foods. CRC. Crit. Rev. Fd. Technol. 8:81.

Giddings, G. C. 1977b. Symposium: the basis of quality in muscle foods. The basis of color muscle foods. J. Food Sci. 42:288.

Gill, J. L. 1978. Design and Analysis of Experiments in Animal and Medical Sciences. Volume 1, 1st ed., p. 135. The Iowa State University Press, Ames, Iowa.

Gordon, A. 1972. Meat and poultry flavor. The Flav. Ind. 3:445.

Gough, T. A., MacPhail, M. F., Webb, K. S., Wood, B. J. and Coleman, R. F. 1977. An examination of some food stuffs for the presence of volatile N-nitrosamines. J. Sci. Fd. Agric. 28:345.

Goutefongea, R., Cassens, R. G. and Woolford, G. 1977. Distribution of sodium nitrite in adipose tissue during curing. J. Food Sci. 42:1637.

Govindarajan, S. 1973. Fresh meat color. CRC Crit. Rev. Fd. Technol. 4:117.

Gray, J. I. 1976. N-nitrosamines and their precursors in bacon: a review. J. Milk Food Technol. 39:686.

Gray, J. I., Bussey, D. M., Dawson, L. E., Price, J. F., Stevenson, K. E., Owens, J. L. and Robach, M. C. 1981a. Investigation into the formation of N-nitrosamines in heated chicken frankfurters. J. Food Sci. (in press).

Gray, J. I. and Collins, M. E. 1977. A comparison of proline and putrescine as precursors of N-nitrosopyrrolidine in nitrite-treated pork system. J. Food Sci. 42:1034.

Gray, J. I. and Collins, M. E. 1978. Formation of N-nitrosopyrrolidine in fried bacon. J. Food Protect. 41:36.

Gray, J. I., Collins, M. E. and MacDonald, B. 1978. Precursors of N-nitrosodimethylamine in fried bacon. J. Food Protect. 41:31.

Gray, J. I., Dawson, L. E., Price, J. F. and Stevenson, K. E. 1979. Nitrites in cured poultry products: chicken frankfurters (in preparation).

Gray, J. I. and Dugan, L. R. 1975. Formation of N-nitrosopyrrolidine from proline and collagen. J. Food Sci. 40:484.

Gray, J. I., MacDonald, B., Pearson, A. M. and Morton, I. D. 1981b. The role of nitrite in cured meat flavor: a review. J. Food Protect. 44:302.

Gray, J. I. and Randall, C. J. 1979. The nitrite-N-nitrosamine problem in meats: an update. J. Food Protect. 42:168.

Greene, B. E. and Price, L. G. 1975. Oxidation-induced color and flavor changes in meat. J. Agr. Food Chem. 23:164.

Guthertz, L. S., Fruin, J. T., Spicer, D. and Fowler, J. L. 1976. Microbiology of fresh comminuted turkey meat. J. Milk Food Technol. 39:823.

Hadden, J. P., Ockerman, H. W., Cahill, V. R., Parrett, N. A. and Barton, R. J. 1975. Influence of sodium nitrite on the chemical and organoleptic properties of comminuted pork. J. Food Sci. 40:626.

Hansen, J. D. and Appleman, M. D. 1955. The effect of sorbic acid, proprionic and caproic acids on the growth of certain Clostridia. Food Res. 20:92.

Hargett, S. M., Blumer, T. N., Hamann, D. D., Keeton, J. T. and Monroe, R. J. 1980. Effect of sodium acid pyrophosphate on sensory, chemical and physical properties of frankfurters. J. Food Sci. 45:905.

Havery, D. C., Kline, D. A., Miletta, E. M., Joe, F. L. and Fazio, T. 1976. Survey of food products for volatile N-nitrosamines. J. Assoc. Off. Anal. Chem. 59:540.

Hayatsu, H., Chung, K. C., Kada, T. and Nakajima, T. 1975. Generation of mutagenic compounds by reaction between sorbic acid and nitrite. Mutation Res. 30:417.

Hill, L. H., Webb, N. B., Moncol, N. D. and Adams, A. T. 1973. Changes in residual nitrite in sausage and luncheon meat products during storage. J. Milk Food Technol. 36:515.

Hirai, C., Herz, K. O., Pokorny, J. and Chang, S. S. 1973. Isolation and identification of volatile flavor compounds in boiled beef. J. Food Sci. 38:393.

Holley, R. A. 1978. Botulism hazard in cured meats treated with low concentrations of nitrite. Report for the Industry/Government Committee on Nitrites and Nitrosamines. Food Research Institute, September 28.

Howard, A., Duffy, P., Else, K. and Brown, W. D. 1973. Possible substitutes for nitrite for pigment formation in cured meat products. J. Agr. Food Chem. 21:894.

Huhtanen, C. N. and Feinberg, J. 1980. Sorbic acid inhibition of Clostridium botulinum in nitrite-free poultry frankfurters. J. Food Sci. 45:453.

Hustad, G. O., Cerveny, J. G., Trenk, H., Deibel, R. H., Kautter, D. A., Fazio, T., Johnston, R. W. and Kolari, O. E. 1973. Effect of sodium nitrite and sodium nitrate on botulinal toxin production and N-nitrosamine formation in weiners. Applied Microbiol. 26:22.

Igene, J. O., King, J. A., Pearson, A. M. and Gray, J. I. 1979. Influence of heme pigments, nitrite and nonheme iron on the development of warmed-over flavor in cooked meat. J. Agr. Food Chem. 27:838.

Ivey, F. J. and Robach, M. C. 1978. Effect of sorbic acid and sodium nitrite on Clostridium botulinum outgrowth and toxin production in canned comminuted pork. J. Food Sci. 43:1782.

Ivey, F. J., Shaver, K. J., Christiansen, L. N. and Tompkin, R. B. 1978. Effect of potassium sorbate on the toxinogenesis by Clostridium botulinum in bacon. J. Food Protect. 41:621.

Jeremiah, L. E., Carpenter, Z. L. and Smith, G. C. 1972. Beef color as related to consumer acceptability and palatability. J. Food Sci. 37:476.

Johnston, M. A., Pivnick, H. and Samson, J. M. 1969. Inhibition of Clostridium botulinum by sodium nitrite in a bacteriological medium and in meat. Can. Institit. Fd. Technol. J. 2:52.

Kanner, J. 1979a. S-Nitrosocysteine (RSNO), an effective antioxidant in cured meat. J. Amer. Oil Chem. Soc. 56:74.

Kanner, J. 1979b. Nitric oxide myoglobin as inhibitor of lipid oxidation. Abstr. 39th Annual IFT Meeting, St. Louis, MO. June, 1979. Paper 22.

Kaul, A., Singh, J. and Kuila, R. K. 1981. Potassium sorbate as a preservative of butter. J. Food Protect. 44:33.

Kemp, J. D. 1974. Nitrate and nitrite substitutes in meat curing. Food Prod. Develop. 8:64.

Kemp, J. D., Fox, J. D. and Moody, W. G. 1974. Cured ham properties as affected by nitrate and nitrite and fresh pork quality. J. Food Sci. 39:972.

Kemp, J. D., Langlois, B. E., Fox, J. D. and Varney, W. Y. 1975. Effects of curing ingredients and holding times and temperatures on the organoleptic and microbiological properties of dry-cured sliced ham. J. Food Sci. 40:634.

Kemp, J. D., Langlois, B. E., Solomon, M. B. and Fox, J. D. 1979. Quality of bonless, dry-cured ham produced with or without nitrate, netting or potassium sorbate. J. Food Sci. 44:914.

Kerr, R. H., Marsy, C. T. N., Schroeder, W. F. and Boyer, E. A. 1926. The use of sodium nitrite in curing meat. J. Agric. Res. 33:541.

Kimoto, W. I., Wasserman, A. E. and Talley, F. B. 1976. Effect of sodium nitrite and sodium chloride on the flavor of processed pork bellies. Lebensm-Wiss. U-Technol. 9:99.

Knowles, M. E., Gilbert, J. and McWeeny, D. J. 1974. Stability of red food colors in the presence of nitrite in canned pork luncheon meat. J. Sci. Fd. Agric. 25:239.

Koizumi, C. and Brown, W. D. 1971. Formation of nitrosylmyoglobin by nicotinamide adenine dinucleotides and flavins. J. Food Sci. 36:1105.

Kraft, A. A. and Ayres, J. C. 1954. Color changes in packaged bologna. Food Technol. 7(1):22.

Kubberod, G., Cassens, R. G. and Greaser, M. L. 1974. Reaction of nitrite with sulfhydryl groups of myosin. J. Food Sci. 39:1228.

Landmann, W. A. and Batzer, O. F. 1966. Influence of processing procedures on chemistry of meat flavor. J. Agr. Food Chem. 14:210.

LaRocco, K. A. and Martin, S. E. 1981. Effects of potassium sorbate alone and in combination with sodium chloride on growth of Salmonella typhimurium 7136. J. Food Sci. 46:568.

Lechowich, R. V., Brown, W. L., Deibel, R. H. and Somers, I. I. 1978. The role of nitrite in the production canned cured meat products. Food Technol. 32(5):45.

Lee, S. H., Cassens, R. G., and Sugiyama, H. 1978. Factors affecting inhibition of Clostridium botulinum in cured meats. J. Food Sci. 43:1371.

Lijinsky, W. and Epstein, S. S. 1970. Nitrosamines as environmental carcinogens. Nature 225:21.

Lillard, D. A. and Ayres, J. C. 1969. Flavor compounds in country-cured hams. Food Technol. 23(2):251.

Lin, H-S. and Sebranek, J. G. 1979. Effect of nitrite concentration and packaging conditions on color stability and rancidity development on sliced bologna. J. Food Sci. 44:1451.

Lin, H-S., Sebranek, J. G., Gallaway, D. E. and Lind, K. D. 1980. Effect of sodium erythorbate and packaging conditions on color stability and rancidity development in sliced bologna. J. Food Sci. 45:115.

Love, J. D. and Pearson, A. M. 1971. Lipid oxidation in meat and meat products--a review. J. Amer. Oil Chem. Soc. 48:547.

Love, J. D. and Pearson, A. M. 1974. Metmyoglobin and nonheme iron as prooxidants in cooked meat. J. Agr. Food Chem. 22:1032.

Love, J. D. and Pearson, A. M. 1976. Metmyoglobin and nonheme iron as prooxidants in egg yolk phospholipid dispersions and cooked meat. J. Agr. Food Chem. 24:494.

MacDonald, B. 1978. The role of nitrite in cured meat flavor. M.S. Thesis, University of Guelph, Guelph.

MacDonald, B., Gray, J. I. and Gibbons, L. N. 1980a. Role of nitrite in cured meat flavor: antioxidant role of nitrite. J. Food Sci. 45:893.

MacDonald, B., Gray, J. I., Kakuda, U. and Lee, M. L. 1980b. Role of nitrite in cured meat flavor: chemical analysis. J. Food Sci. 45:889.

MacDonald, B., Gray, J. I., Stanley, D. W. and Usborne, W. R. 1980c. Role of nitrite in cured meat flavor: sensory analysis. J. Food Sci. 45:885.

MacDougall, D. B., Mottram, D. S. and Rhodes, D. N. 1975. Contribution of nitrite and nitrate to the colour and flavour of cured meats. J. Sci. Fd. Agric. 26:1743.

MacNeil, J. H., Dimick, P. S. and Mast, M. G. 1973. Use of chemical compounds and a rosemary spice extract in quality maintenance of deboned poultry meat. J. Food Sci. 38:1080.

MacNeil, J. H. and Mast, M. G. 1973. Frankfurters without nitrate or nitrite. Food Prod. Develop. 7:36.

Magee, P, B, and Barnes, J. M. 1956. The production of malignant primary hepatic tumours in the rat by feeding dimethylnitrosamine. Brit. J. Cancer 10:114.

McKinley, G. A. and Avens, J. S. 1981. Microbial quality of ground and comminuted, raw and cooked turkey meat. J. Food Protect. 44:139.

Melnick, D., Luckmann, F. H. and Gooding, C. M. 1954a. Sorbic acid as a fungistatic agent for foods. V. Resistance of sorbic acid in cheese to oxidative deterioration. Food Res. 19:33.

Melnick, D., Luckmann, F. H. and Gooding, C. M. 1954b. Sorbic acid as a fungistatic agent for foods. VI. Metabolic degradation of sorbic acid in cheese by molds and mechanism of mold inhibition. Food Res. 19:44.

Mirvish, S. S., Wallcave, L., Eager, M. and Shubik, P. 1972. Ascorbate-nitrite reaction: possible means of blocking the formation of carcinogenic N-nitroso compounds. Science 177:65.

Mottram, D. S. and Patterson, R. L. S. 1977. The effect of ascorbate reductants on N-nitrosamine formation in a model system resembling bacon fat. J. Sci. Fd. Agric. 28:352.

Mottram, D. S., Patterson, R. L. S., Rhodes, D. N. and Gough, T. A. 1975. Influence of ascorbic acid and pH on the formation of N-nitroso-dimethylamine in cured pork containing added dimethylamine. J. Sci. Fd. Agric. 26:47.

Nakamura, M., Baba, N., Nakaoka, T., Wada, Y., Ishibashi, T. and Kawabata, T. 1976. Pathways of formation of N-nitrosopyrrolidine in fried bacon. J. Food Sci. 41:874.

Namiki, M. and Kada, T. 1975. Formation ethylnitrolic acid by reaction of sorbic acid with sodium nitrite. Agr. Biol. Chem. 39:1135.

Namiki, M., Osawa, T., Ishibashi, H., Namiki, K. and Tsuiji, K. 1981. Chemical aspects of mutagen formation by sorbic acid-sodium nitrite reaction. J. Agr. Food Chem. 29:407.

Newberne, P. M. 1979. Nitrite promotes lymphoma incidence in rats. Science 204:1079.

Nitrite Safety Council. 1980. A survey of N-nitrosamines in sausages and dry-cured meat products. Food Technol. 34(7):45.

Nordin, H. R. 1969. The depletion of added sodium nitrite in ham. Can. Instit. Fd. Technol. 2:79.

Ockerman, H. W., Blumer, T. N. and Craig, H. B. 1964. Volatile chemical compounds in dry-cured hams. J. Food Sci. 29:123.

Olson, V. M., King, N. A., Langbelm, J. A. and Stadelman, W. J. 1979. Acceptability of smoked turkey drumsticks with and without nitrite addition. Poultry Sci. 58:587.

Paquette, M. W., Robach, M. C., Sofos, J. N. and Busta, F. F. 1980. Effect of various concentrations of sodium nitrite and potassium sorbate on color and sensory quality of bacon. J. Food Sci. 45:1293.

Pearson, A. M., Love, J. D. and Shorland, F. B. 1977. Warmed-over flavor in meat, poultry and fish. Adv. Food Res. 23:1.

Pensabene, J. W., Feinberg, J. I., Dooley, C. J., Phillips, J. G. and Fiddler, W. 1979. Effect of pork belly composition and nitrite level on nitrosamine formation in fried bacon. J. Agr. Food Chem. 27:842.

Pensabene, J. W., Fiddler, W., Gates, R. A., Fagen, J. C. and Wasserman, A. E. 1974. Effect of frying and other cooking conditions on N-nitrosopyrrolidine formation in bacon. J. Food Sci. 39:314.

Pensabene, J. W., Fiddler, W., Mergens, W. and Wasserman, A. E. 1978. Effect of α-tocopherol formation on the inhibition of N-nitrosopyrrolidine formation in model systems. J. Food Sci. 43:80.

Persson, T. and von Sydow, E. 1973. Aroma of canned beef: gas chromatographic and mass spectrometric analysis of the volatiles. J. Food Sci. 38:377.

Persson, T., von Sydow, E. and Akesson, C. 1973. Aroma of canned beef: sensory properties. J. Food Sci. 38:386.

Pierson, M. D., Smoot, L. A. and Stern, N. J. 1979. Effect of potassium sorbate on growth of Staphylococcus aureus in bacon. J. Food Protect. 42:302.

Piotrowski, E. G., Zaika, L. L. and Wasserman, A. E. 1970. Studies on aroma of cured ham. J. Food Sci. 35:321.

Price, L. G. and Greene, B. E. 1978. Factors affecting panelist's perceptions of cured meat flavor. J. Food Sci. 43:319.

Price, J. F. and Stevenson, K. E. 1979. Effect of sorbate and nitrite in bacon on color, flavor and Clostridium botulinum toxinogenesis. Abstr. 39th Annual IFT Meeting, St. Louis, MO., June, 1979. Paper 372.

Reineccius, G. A. 1979. Symposium on meat flavor. Off-flavor in meat and fish--a review. J. Food Sci. 44:12.

Reith, J. F. and Szakaly, M. 1967a. Formation and stability of nitrosylmyoglobin. 1. Studies with model systems. J. Food Sci. 32:188.

Reith, J. F. and Szakaly, M. 1967b. Formation and stability of nitrosylmyoglobin. 11. Studies on meat. J. Food Sci. 32:194.

Renerre, M. and Rougie, P. 1979. Influence du chauffage sur la fixation du nitrite a la myoglobine. Ann. Technol. Agric. 28:423.

Rhodes, A. C. and Jarvis, B. 1976. A pork slurry system for studying inhibition of Clostridium botulinum by curing salts. J. Fd. Technol. 11:13.

Riemann, H., Lee, W. H. and Genigeorgis, C. 1972. Control of Clostridium botulinum and Staphylococcus aureus in semi-preserved meat products. J. Milk Food Technol. 35:514.

Robach, M. C. and Adams, K. 1979. Human reaction related to sensory evaluation of cooked bacon. Monsanto Ind. Chem. Co., St. Louis, MO. October, 1979.

Robach, M. C., DiFate, V. G., Adams, K. and Kier, L. D. 1980a. Evaluation of the nutagenicity of sorbic acid-sodium nitrite reaction products produced in bacon-curing brines. Fd. Cosmet. Toxicol. 18:237.

Robach, M. C. and Ivey, F. J. 1978. Antimicrobial efficacy of potassium sorbate dip in freshly processed poultry. J. Food Protect. 41:284.

Robach, M. C., Ivey, F. J. and Hickey, C. S. 1978. System for evaluating Clostridia inhibition in cured meat products. Applied Environ. Microbiol. 36:210.

Robach, M. C., To, E. C., Meydav, S. and Cook, C. F. 1980b. Effects of sorbate on microbial growth in cooked turkey products. J. Food Sci. 45:638.

Robach, M. C., Owens, J. L., Paquette, M. W., Sofos, J. N. and Busta, F. F. 1980c. Effects of various concentrations of sodium nitrite and potassium sorbate on nitrosamine formation in commercially prepared bacon. J. Food Sci. 45:1280.

Roberts, T. A. 1975. The microbiological role of nitrate and nitrite. J. Sci. Fd. Agric. 26:1755.

Sales, C. A., Bowers, J. A. and Kropf, D. 1980. Consumer acceptability of turkey franks with 0, 40 and 100 ppm nitrite. J. Food Sci. 45:1060.

Sanderson, A., Pearson, A. M. and Schweigert, B. S. 1966. Effect of cooking procedures and flavor components of beef carbonyl components. J. Agr. Food Chem. 14:245.

Sato, K. and Hegarty, G. R. 1971. Warmed-over flavor in cooked meat. J. Food Sci. 36:1098.

Scanlan, R. A. 1975. N-nitrosamines in foods. CRC. Crit. Rev. Fd. Technol. 5:357.

Sebranek, J. G. 1979. Advances in the technology of nitrite use and consideration of alternatives. Food Technol. 33(7):58.

Sebranek, J. G. and Cassens, R. G. 1973. Nitrosamines--a review. J. Milk Food Technol. 36:76.

Sebranek, J. G., Cassens, R. G., Hoekstra, W. G., Winder, W. C., Podebradsky, E. V. and Kielsmeier, E. W. 1973. $^{15}N$ tracer studies of nitrite added to a comminuted meat product. J. Food Sci. 38:1220.

Sebranek, J. G., Schroder, B. G., Rust, R. E. and Topel, D. G. 1977. Influence of sodium erythorbate on color development, flavor and overall acceptability of frankfurters cured with reduced levels of sodium nitrite. J. Food Sci. 42:1120.

Sen, N. P., Donaldson, B., Iyengar, J. R. and Panalaks, T. 1973. N-nitrosopyrrolidine and N-nitrosodimethylamine in bacon. Nature 241:473.

Sen, N. P., Donaldson, B., Seaman, S., Iyengar, J. R. and Miles, W. F. 1976. Inhibition of N-nitrosamines in fried bacon by propyl gallate and L-ascorbyl palmitate. J. Agr. Food Chem. 24:397.

Sen, N. P., Seaman, S. and Miles, W. F. 1979. Volatile N-nitrosamines in various cured meat products: effect of cooking and recent trends. J. Agr. Food Chem. 27:1354.

Sen, N. P., Iyengar, J. R., Donaldson, B. A. and Panalaks, T. 1974. Effect of sodium nitrite concentration on formation of N-nitroso-pyrrolidine and N-nitrosodimethylamine in fried bacon. J. Agr. Food Chem. 22:540.

Shank, R. C. and Newberne, P. M. 1976. Dose-response study of the carcinogenicity of dietary sodium nitrite and morpholine in rats and hamsters. Fd. Cosmet. Toxicol. 14:1.

Shibamoto, T., Kamiya, Y. and Mihara, S. 1981. Isolation and identification of volatile compounds in cooked meats: sukiyaki. J. Agr. Food Chem. 29:57.

Shults, G. W., Cohen, J. S., Howker, J. J. and Wierbicki, E. 1977. Effects of sodium nitrate and sodium nitrite addition and irradiation process variables on the color and acceptability of corned beef briskets. J. Food Sci. 42:1506.

Siedler, A. J. and Schweigert, B. S. 1959. Effects of heat, nitrite level, ferrous salts and reducing agents on formation of denatured nitrosylmyoglobin. J. Agr. Food Chem. 7:271.

Simon, S., Ellis, D. E., MacDonald, B. D., Miller, D. G., Waldman, R. C. and Werterberg, D. O. 1973. Influence of nitrite and nitrate ingredients on quality of packaged frankfurters. J. Food Sci. 38:919.

Sink, J. D. 1979. Symposium on meat flavor. Factors influencing the flavor of muscle foods. J. Food Sci. 44:1.

Skjelkvale, R. and Tjaberg, T. B. 1974. Comparison of salami sausage products with and without addition of nitrite and nitrate. J. Food Sci. 39:520.

Smith, D. P. and Rollin, N. J. 1954. Sorbic acid as a fungistatic agent for foods. VII. Effectiveness of sorbic acid in protecting cheese. Food Res. 19:59.

Sofos, J. N. and Busta, F. F. 1980. Alternatives to the use of nitrite as an antibotulinal agent. Food Technol. 34(5):244.

Sofos, J. N., Busta, F. F. and Allen, C. E. 1979a. Botulism control by nitrite and sorbate in cured meats: a review. J. Food Protect. 42:739.

Sofos, J. N., Busta, F. F. and Allen, C. E. 1979b. Sodium nitrite and sorbate effects on Clostridium botulinum spore germination and total microbial growth in chicken frankfurter emulsion during temperature abuse. Applied Environ. Microbiol. 37:1103.

Sofos, J. N., Busta, F. F. and Allen, C. E. 1980a. Influence of pH on Clostridium botulinum control by sodium nitrite and sorbic acid in chicken emulsions. J. Food Sci. 45:7.

Sofos, J. N., Busta, F. F., Bhothipaksa, K. and Allen, C. E. 1979c. Sodium nitrite and sorbate effects on Clostridium botulinum toxin formation in chicken frankfurter-type emulsion. J. Food Sci. 44:668.

Sofos, J. N., Busta, F. F., Bhothipaksa, K., Allen, C. E., Robach, M. C. and Paquette, M. W. 1980b. Effects of various concentrations of sodium nitrite and potassium sorbate on Clostridium botulinum toxin production in commercially prepared bacon. J. Food Sci. 45:1285.

Stone, H., Sidel, J., Oliver, S., Woolsey, A. and Singleton, R. C. 1974. Sensory evaluation by quantitative descriptive analysis. Food Technol. 28(11):24.

Swain, J. W. 1972. Volatile flavor constituents of pork cured with and without nitrite. PhD. Dissertation, University of Missouri, Columbia.

Tanaka, K., Chung, K. C., Hayatsu, H. and Kada, T. 1978. Inhibition of nitrosamine formation in-vitro by sorbic acid. Fd. Cosmet. Toxicol. 16:209.

Tanaka, N., Traisman, E., Lee, M. H., Cassens, R. G. and Foster, E. M. 1980. Inhibition of botulinal toxin formation in bacon by acid development. J. Food Protect. 43:450.

Tarladgis, B. G. 1962. Interpretation of spectra of meat pigments. 11. Cured meats. The mechanism of colour fading. J. Sci. Fd. Agric. 13:485.

Tarladgis, B., Watts, B. M., Younathan, M. T. and Dugan, L. R. 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. J. Amer. Oil Chem. Soc. 37:44.

Tims, M. J. and Watts, B. M. 1958. Protection of cooked meats with phosphates. Food Technol. 12:240.

Tompkin, R. B. 1980. Botulism from meat and poultry products--a historical perspective. Food Technol. 34(5):229.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1977. Variation in inhibition of Clostridium botulinum by nitrite in perishable canned comminuted cured meat. J. Food Sci. 42:1046.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1978a. Enhancing nitrite inhibition of Clostridium botulinum with isoascorbate in perishable canned cured meat. Applied Environ. Microbiol. 35:59.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1978b. Effect of prior refrigeration on botulinal growth in perishable canned cured meat when temperature abused. Applied Environ. Microbiol. 35:863.

Tompkin, R. B., Christiansen, L. N. and Shaparis, R. B. 1978c. Causes of variation in botulinal inhibition in perishable canned cured meat. Applied Environ. Microbiol. 35:886.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1978d. Antibotulinal role of isoascorbate in cured meat. J. Food Sci. 43:1368.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1978e. The effect of iron on botulinal inhibition in perishable canned cured meat. J. Fd. Technol. 13:521.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1979a. Iron and the antibotulinal efficacy of nitrite. Applied Environ. Microbiol. 37:351.

Tompkin, R. B., Christiansen, L. N. and Shaparis, A. B. 1979b. Isoascorbate level and botulinal inhibition in perishable canned cured meat. J. Food Sci. 44:1147.

Uebersax, M. A., Dawson, L. E. and Uebersax, K. L. 1978. Storage stability (TBA) of meat obtained from turkeys receiving tocopherol supplementation. Poultry Sci. 57:937.

Uram, G., Carpenter, J. A. and Reagen, J. O. 1981. Effect of particle size, casing diameter and addition of emulsions and residual nitrite on quality of cooked salami. J. Food Sci. 46:842.

U.S.D.A. 1979. Final report. Shelflife, sensory, cookery and physical characteristics of bacon cured with varying levels of sodium nitrite and potassium sorbate. Meat Science Research Laboratory, Beltsville, MD. July, 1979.

von Elbe, J. H., Klement, J. T., Amundsen, C. H., Cassens, R. G. and Lindsay, R. C. 1974. Evaluation of betalain pigments as substitute sausage colorants. J. Food Sci. 39:128.

Waldman, R. C., Westerberg, D. O. and Simon, S. 1974. Influence of curing ingredients and storage time on quality of preblended sausage meats and frankfurters. J. Food Sci. 39:718.

Walsh, K. A. and Rose, D. 1956. Factors affecting oxidation of nitric oxide myoglobin. J. Agr. Food Chem. 4:352.

Walters, C. L. and Taylor, A. M. 1963. Biochemical properties of pork muscle in relation to curing. Food Technol. 17(3):354.

Wasserman, A. E. 1979. Symposium on meat flavor. Chemical basis for meat flavor: a review. J. Food Sci. 44:6.

Wasserman, A. E. and Fiddler, R. N. 1976. Processing and sensory evaluation of a primal cut cured with nitrite alone or with nitrite-nitrate mixture. Eastern Regional Research Center. Philadelphia, PA. June 4, 1976.

Wasserman, A. E., Kimoto, W. and Phillips, J. G. 1977. Consumer acceptance of nitrite-free bacon. J. Food Protect. 40:683.

Wasserman, A. E. and Talley, F. 1972. The effect of sodium nitrite on the flavor of frankfurters. J. Food Sci.. 37:536.

Watts, B. M. and Lehmann, B. T. 1965. The effect of ascorbic acid on the oxidation of hemoglobin and the formation of nitrosylhemoglobin. Food Res. 17:100.

Weiss, T. J., Green, R. and Watts, B. M. 1953. Effect of metal ions on the formation of nitrosylhemoglobin. Food Res. 18:11.

Williams, D. L. H. 1978. Comparison of the efficiencies of ascorbic and sulphamic acid as nitrite traps. Fd. Cosmet. Toxicol. 16:365.

Williams, J. C. and Greene, B. E. 1979. Plate waste of bacon cured with and without sodium nitrite. J. Food Sci. 44:1260.

Wilson, B. R., Pearson, A. M. and Shorland, F. B. 1976. Effect of total lipids and phospholipids on warmed-over flavor of red and white muscle from several species as measured by TBA analysis. J. Agr. Food Chem. 24:7.

Wolff, I. A. and Wasserman, A. E. 1972. Nitrate, nitrite and nitrosamines. Science 177:15.

Woolford, G. and Cassens, R. G. 1977. The fate of sodium nitrite in bacon. J. Food Sci. 42:586.

Woolford, G., Cassens, R. G., Greaser, M. L. and Sebranek, J. G. 1976. The fate of nitrite: reaction with protein. J. Food Sci. 41:585.

Yarbrough, J. M., Rake, J. B. and Eagon, R. G. 1980. Bacterial inhibition effects of nitrite: inhibition of active transport, but not of group translocation and intracellular enzymes. Applied Environ. Microbiol. 39:831.

York, G. K. and Vaughn, R. H. 1954. Use of sorbic acid enrichment media for species of Clostridium. J. Bacteriol. 68:739.

York, G. K. and Vaughn, R. H. 1955. Resistance of Clostridium botulinum to sorbic acid. Food Res. 20:60.

Younathan, M. T. and Watts, B. M. 1959. Relationship of meat pigments to lipid oxidation. Food Res. 24:728.

Zipser, M. W., Kwon, T-W. and Watts, B. M. 1964. Oxidative changes in cured and uncured frozen cooked pork. J. Agr. Food Chem. 12:105.

Zipser, M. W. and Watts, B. M. 1962. A modified 2-thiobarbituric acid (TBA) method for the determination of malonaldehyde in cured meats. Food Technol. 16(7):102.

# APPENDIX

# APPENDIX

## SENSORY PANEL EVALUATION FORM

Name _____ Date _____

This sensory evaluation panel will evaluate the odor, color and flavor of various hams. Compare each sample to the control sample (R) on your tray.

Answer each question in the proper sequence, placing a vertical line across the horizontal line at the point that best describes that property in the sample. Take sufficient sample and time to evaluate each characteristic.

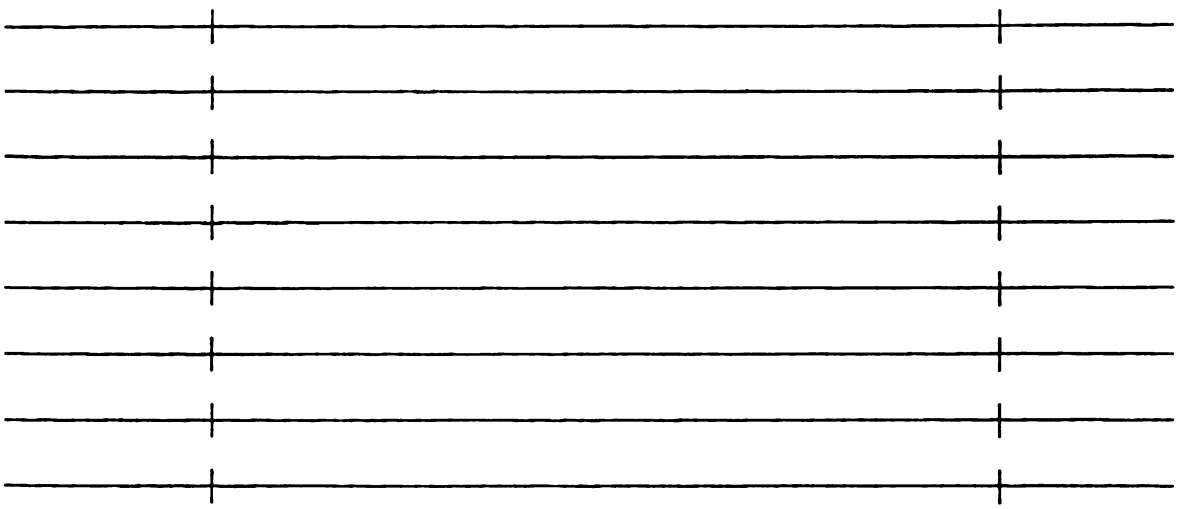
Please make sure that you clearly label each vertical line with the proper sample number at the time of evaluation. (Please label a vertical line for (R) also.)

Water will be supplied for rinsing between samples.

Please evaluate the samples for color, comparing each sample to the control sample (R) on your tray. Please label each vertical line with proper sample number--including (R) also.

HAM COLOR:

Unacceptable Acceptable



You are receiving samples of ham to compare for odor.

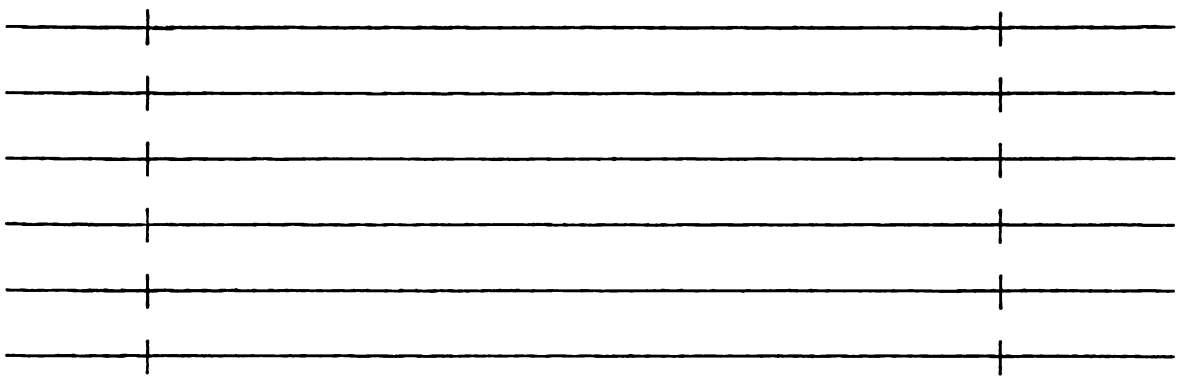
AROMA: Remove cover over sample.

While holding each dish close to your nose, take 3 short sniffs.

Please label each vertical line with the proper sample number - including (R).

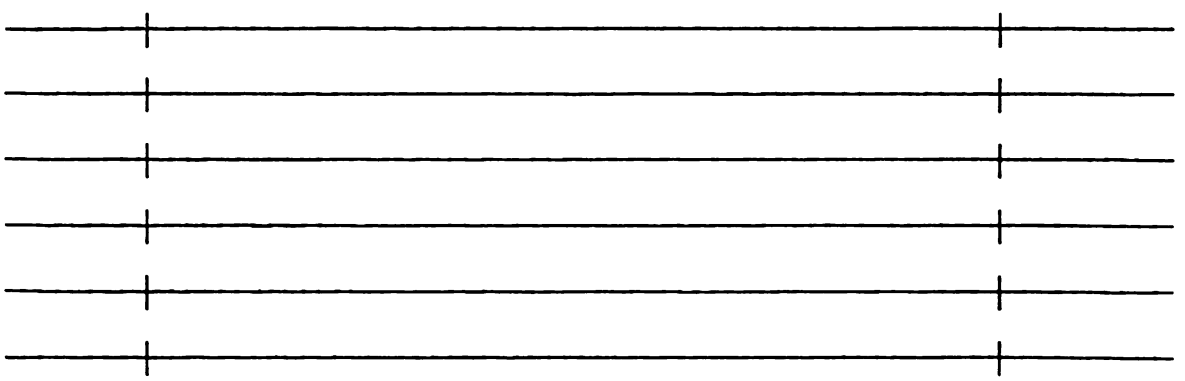
HAM ODOR:

Weak Strong



OFF-ODORS:

Weak Strong

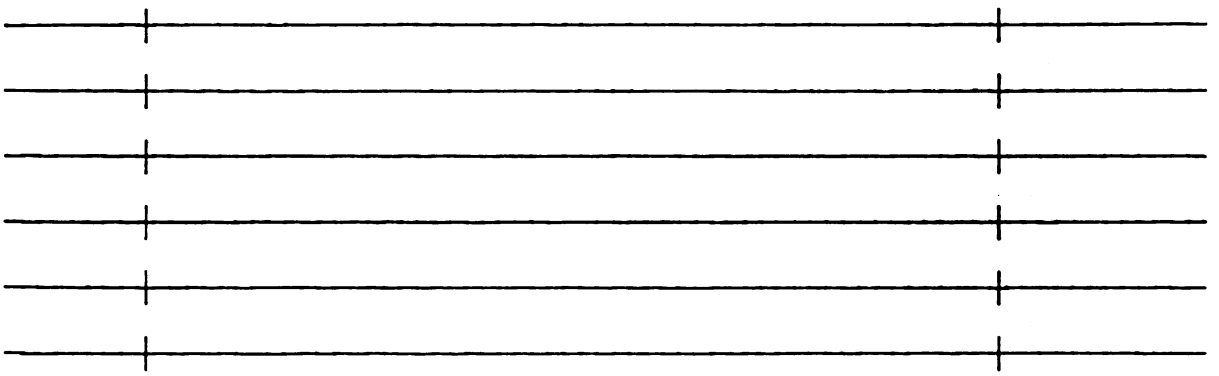


FLAVOR _ Chew each sample of meat normally. Perceive total flavor on tongue. You do not need to swallow it. Refer to reference (R) as often as necessary.

- Rinse your mouth between each sample.

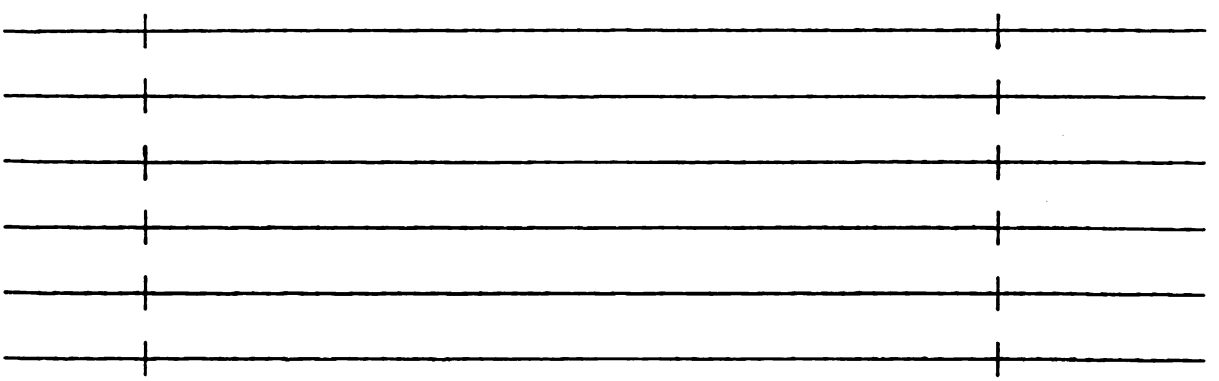
HAM FLAVOR:

Weak Strong



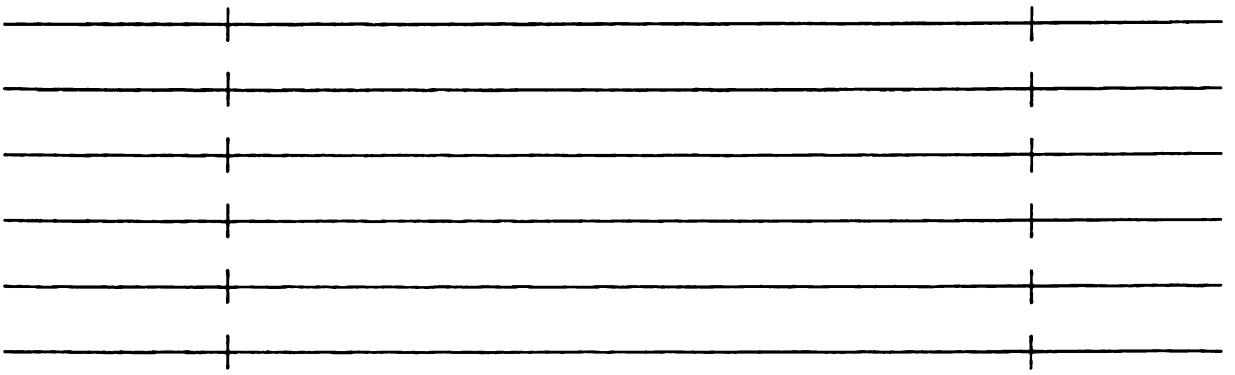
OFF-FLAVOR:

Weak Strong



OVERALL SATISFACTION

Unacceptable Acceptable



Any further comments:

Thank-you for your participation.

# APPENDIX

## TABLES A-1 THROUGH A-64

Table A-1: Analysis of variance on the mean sensory scores for turkey bologna color.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 95.88 | 1.87 |
| Treatments (B) | 5 | 20479.40 | 1059.90** |
| Time (C) | 3 | 153.87 | 0.78 |
| A X B | 95 | 19.32 | 0.38 |
| A X C | 57 | 197.27 | 3.85 |
| B X C | 15 | 57.00 | 1.87 |
| A X B X C | 285 | 30.41 | 0.59 |
| Error | 480 | 51.23 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-2: A comparison between treatment means for turkey bologna color using Dunnett's Test ($p < 0.01$).

| Treatments (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 1.72 | 9.89 | 10.42 | 10.56 | 10.41 | 10.94 |

Table A-3: Analysis of variance on the mean sensory scores for turkey bologna flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 34.17 | 3.71 |
| Treatments (B) | 5 | 1008.99 | 236.90** |
| Time (C) | 3 | 25.34 | 0.79 |
| A X B | 95 | 4.26 | 0.46 |
| A X C | 57 | 31.70 | 3.45 |
| B X C | 15 | 6.29 | 1.09 |
| A X B X C | 285 | 5.79 | 0.63 |
| Error | 480 | 9.20 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-4: Analysis of variance on the mean sensory scores for turkey bologna odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 385.05 | 4.47 |
| Treatments (B) | 5 | 10086.30 | 211.50** |
| Time (C) | 3 | 29.03 | 0.10 |
| A X B | 95 | 47.69 | 0.55 |
| A X C | 57 | 292.95 | 3.39 |
| A X B X C | 285 | 49.85 | 0.58 |
| Error | 480 | 86.11 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-5: A comparison of treatment means for turkey bologna flavor using Dunnett's Test ($p < 0.01$).

| Treatments (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 2.50 | 8.14 | 8.38 | 8.38 | 8.41 | 9.43 |

Table A-6: A comparison of treatment means for turkey bologna odor using Dunnett's Test ($p < 0.01$).

| Treatments (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 3.19 | 9.14 | 9.18 | 9.21 | 9.02 | 9.91 |

Table A-7: Analysis of variance on the mean sensory scores for turkey bologna off-flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 29.63 | 3.27 |
| Treatments (B) | 5 | 360.93 | 11.74** |
| Time (C) | 3 | 15.15 | 0.57 |
| A X B | 95 | 30.74 | 3.39 |
| A X C | 57 | 26.61 | 2.93 |
| B X C | 15 | 5.27 | 0.69 |
| A X B X C | 285 | 7.65 | 0.84 |
| Error | 480 | 9.07 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-8: Analysis of variance on the mean sensory scores for turkey bologna off-odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 39.53 | 4.29 |
| Treatments (B) | 5 | 792.01 | 118.92** |
| Time (C) | 3 | 32.68 | 0.98 |
| A X B | 95 | 6.66 | 0.72 |
| A X C | 57 | 33.42 | 3.63 |
| B X C | 15 | 5.78 | 1.27 |
| A X B X C | 285 | 4.55 | 0.49 |
| Error | 480 | 9.20 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-9: A comparison of treatment means for turkey bologna off-flavor using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 4.24 | 5.81 | 2.29 | 2.21 | 2.29 | 2.32 |

Table A-10: A comparison of treatment means for turkey bologna off-flavor using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 7.53 | 2.20 | 2.02 | 2.23 | 2.25 | 1.78 |

Table A-11: Analysis of variance on the mean sensory scores for the overall acceptability of turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 19 | 184.87 | 2.63 |
| Treatments (B) | 5 | 15639.30 | 348.30** |
| Time (C) | 3 | 310.93 | 1.07 |
| A X B | 95 | 44.90 | 0.64 |
| A X C | 57 | 291.64 | 4.15 |
| B X C | 15 | 80.20 | 1.39 |
| A X B X C | 285 | 57.42 | 0.82 |
| Error | 480 | 70.32 | |
| Total | 959 | | |

**($p < 0.01$)

Table A-12: Comparison of treatment means for overall acceptability of turkey bologna using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 2.02 | 9.41 | 9.64 | 9.43 | 9.34 | 10.32 |

Table A-13: Analysis of variance on Hunter Color L, $a_L$ and $b_L$ values for turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| L Values | | | |
| Time (A) | 3 | 1.67 | 41.80** |
| Treatment (B) | 5 | 12.17 | 304.25** |
| A X B | 15 | 0.14 | 3.50** |
| Error | 72 | 0.04 | |
| Total | 95 | | |
| $a_L$ Values | | | |
| Time (A) | 3 | 7.53 | 753.00** |
| Treatment (B) | 5 | 21.76 | 2176.00** |
| A X B | 15 | 0.17 | 17.00** |
| Error | 72 | 0.01 | |
| Total | 95 | | |
| $b_L$ Values | | | |
| Time (A) | 3 | 0.59 | 59.00** |
| Treatment (B) | 5 | 14.70 | 1470.00** |
| A X B | 15 | 0.10 | 10.00** |
| Error | 72 | 0.01 | |
| Total | 95 | | |

**($p < 0.01$)

Table A-14: A comparison of treatment means for Hunter Color L, $a_L$ and $b_L$ values of turkey bologna using Dunnett's Test ($p < 0.01$).

| Time | Treatments | | | | | |
|---|---|---|---|---|---|---|
| | 00 | 20 | 40 | 60 | 100 | 156 |
| **L Values** | | | | | | |
| Week 0 | 63.2 | 61.1 | 61.3 | 61.0 | 61.7 | 61.3 |
| Week 1 | 63.4 | 61.3 | 61.5 | 60.9 | 61.4 | 61.5 |
| Week 2 | 63.3 | 60.7 | 61.1 | 61.3 | 61.5 | 61.3 |
| Week 3 | 62.9 | 60.1 | 60.8 | 60.9 | 60.9 | 60.9 |
| **$a_L$ Values** | | | | | | |
| Week 0 | 2.6 | 5.5 | 6.2 | 5.6 | 6.1 | 6.1 |
| Week 1 | 3.8 | 6.6 | 6.8 | 6.6 | 6.8 | 6.8 |
| Week 2 | 4.3 | 6.6 | 6.8 | 6.9 | 7.0 | 6.7 |
| Week 3 | 4.5 | 6.9 | 7.1 | 7.2 | 7.1 | 7.1 |
| **$b_L$ Values** | | | | | | |
| Week 0 | 8.7 | 6.3 | 6.4 | 6.1 | 6.6 | 6.4 |
| Week 1 | 8.6 | 6.1 | 6.2 | 6.4 | 6.2 | 6.4 |
| Week 2 | 8.4 | 5.8 | 6.1 | 6.2 | 6.3 | 6.1 |
| Week 3 | 8.3 | 5.5 | 6.1 | 6.1 | 6.1 | 6.4 |

Table A-15: Analysis of variance on TBA values for turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| Time (A) | 3 | 4.62 | 23.10 ** |
| Treatment (B) | 5 | 137.73 | 688.60 ** |
| A X B | 15 | 3.01 | 15.05 ** |
| Error | 168 | 0.20 | |
| Total | 191 | | |

**($p < 0.01$)

Table A-16: A comparison of treatment means for TBA values for turkey bologna using Dunnett's Test ($p < 0.01$).

| Time | Treatments | | | | | |
|---|---|---|---|---|---|---|
| | 00 | 20 | 40 | 60 | 100 | 156 |
| Week 0 | 5.9 | 2.2 | 2.2 | 2.6 | 1.4 | 1.4 |
| Week 1 | 5.5 | 2.2 | 1.7 | 1.7 | 1.5 | 1.5 |
| Week 2 | 8.0 | 2.3 | 1.8 | 2.5 | 1.8 | 2.1 |
| Week 3 | 8.3 | 1.9 | 1.6 | 1.6 | 1.7 | 1.6 |

Table A-17: Analysis of variance on the mean sensory scores for turkey bologna color.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 9.87 | 1.66 |
| Treatments (B) | 7 | 680.12 | 106.60** |
| Time (C) | 3 | 9.28 | 1.04 |
| A X B | 63 | 6.38 | 1.07 |
| A X C | 27 | 8.96 | 1.51 |
| B X C | 21 | 5.84 | 0.98 |
| A X B X C | 189 | 5.95 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-18: A comparison of treatment means for turkey bologna color using Dunnett's Test ($p < 0.01$).

| Treatment | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 2.77 | 2.20 | 1.76 | 9.70 | 10.41 | 10.26 | 10.30 | 10.27 |

Table A-19: Analysis of variance on the mean sensory scores for turkey bologna flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 22.15 | 2.41 |
| Treatments (B) | 7 | 252.11 | 25.36** |
| Time (C) | 3 | 0.55 | 0.04 |
| A X B | 63 | 9.94 | 1.08 |
| A X C | 27 | 14.62 | 1.59 |
| B X C | 21 | 7.12 | 0.77 |
| A X B X C | 189 | 9.20 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-20: Analysis of variance on the mean sensory scores for turkey bologna odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 19.35 | 2.16 |
| Treatments (B) | 7 | 225.00 | 20.96** |
| Time (C) | 3 | 17.30 | 1.65 |
| A X B | 63 | 10.73 | 1.20 |
| A X C | 27 | 10.49 | 1.17 |
| B X C | 21 | 19.23 | 2.15 |
| A X B X C | 189 | 8.95 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-21: A comparison of treatment means for turkey bologna flavor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 3.60 | 4.02 | 3.64 | 8.59 | 9.03 | 7.94 | 8.00 | 9.09 |

Table A-22: A comparison of treatment means for turkey bologna odor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 4.32 | 5.07 | 4.70 | 8.36 | 9.22 | 9.45 | 9.40 | 9.59 |

Table A-23: Analysis of variance on the mean sensory scores for turkey bologna off-flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 7.75 | 0.76 |
| Treatments (B) | 7 | 251.83 | 35.03** |
| Time (C) | 3 | 10.19 | 0.88 |
| A X B | 63 | 7.19 | 0.70 |
| A X C | 27 | 11.64 | 1.14 |
| B X C | 21 | 11.43 | 1.12 |
| A X B X C | 189 | 10.21 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-24: Analysis of variance on the mean sensory scores for turkey bologna off-odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 16.14 | 1.37 |
| Treatments (B) | 7 | 181.72 | 13.34** |
| Time (C) | 3 | 58.50 | 6.13 |
| A X B | 63 | 13.62 | 1.15 |
| A X C | 27 | 9.54 | 0.81 |
| B X C | 21 | 14.46 | 1.22 |
| A X B X C | 189 | 11.81 | |
| Total | 319 | | |

Table A-25: A comparison of treatment means for turkey bologna off-flavor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 6.75 | 5.50 | 7.25 | 1.49 | 1.62 | 1.74 | 2.51 | 1.57 |

Table A-26: A comparison of treatment means for turkey bologna off-flavor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 6.32 | 6.00 | 6.73 | 2.64 | 2.70 | 1.67 | 2.66 | 1.89 |

Table A-27: Analysis of variance on the mean sensory scores for overall acceptability of turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 11.72 | 1.05 |
| Treatments (B) | 7 | 396.13 | 42.05** |
| Time (C) | 3 | 41.10 | 2.45 |
| A X B | 63 | 9.42 | 0.84 |
| A X C | 27 | 16.79 | 1.51 |
| B X C | 21 | 9.06 | 0.81 |
| A X B X C | 189 | 11.17 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-28: A comparison of treatment means for overall acceptability of turkey bologna using Dunnett's Test ($p < 0.01$)

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 2.31 | 4.39 | 2.70 | 8.90 | 9.28 | 8.43 | 8.52 | 10.03 |

Table A-29: Analysis of variance on Hunter Color L, $a_L$ and $b_L$ values for turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| L Values | | | |
| Time (A) | 3 | 0.54 | 13.50** |
| Treatment (B) | 7 | 1.84 | 46.00** |
| A X B | 21 | 0.48 | 11.50** |
| Error | 96 | 0.04 | |
| Total | 127 | | |
| $a_L$ Values | | | |
| Time (A) | 3 | 5.04 | 252.00** |
| Treatment (B) | 7 | 27.95 | 1397.50** |
| A X B | 21 | 0.07 | 3.50** |
| Error | 96 | 0.02 | |
| Total | 127 | | |
| $b_L$ Values | | | |
| Time (A) | 3 | 0.16 | 8.00** |
| Treatment (B) | 7 | 17.71 | 885.50** |
| A X B | 21 | 0.06 | 3.00** |
| Error | 96 | 0.02 | |
| Total | 127 | | |

**($p < 0.01$)

Table A-30: A comparison of treatment means for Hunter Color L, $a_L$ and $b_L$ values of turkey bologna using Dunnett's Test ($p < 0.01$).

| Time | Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| **L Values** | | | | | | | | |
| Week 0 | 66.8 | 66.4 | 66.5 | 65.4 | 65.6 | 65.4 | 65.4 | 65.4 |
| Week 1 | 66.4 | 65.5 | 65.9 | 65.4 | 65.3 | 65.0 | 65.8 | 65.8 |
| Week 2 | 66.3 | 66.5 | 65.9 | 65.8 | 65.4 | 65.1 | 65.3 | 65.3 |
| Week 3 | 65.7 | 65.9 | 65.7 | 65.5 | 65.6 | 65.9 | 65.9 | 65.9 |
| **$a_L$ Values** | | | | | | | | |
| Week 0 | 2.9 | 2.9 | 2.9 | 5.9 | 6.0 | 5.3 | 5.8 | 5.7 |
| Week 1 | 3.9 | 3.7 | 3.9 | 6.5 | 6.4 | 6.1 | 6.6 | 6.4 |
| Week 2 | 3.3 | 3.3 | 3.3 | 5.9 | 5.9 | 5.3 | 5.8 | 5.8 |
| Week 3 | 3.0 | 3.1 | 3.0 | 5.4 | 5.5 | 5.2 | 5.6 | 5.4 |
| **$b_L$ Values** | | | | | | | | |
| Week 0 | 9.6 | 9.4 | 9.7 | 7.4 | 7.5 | 7.6 | 7.7 | 7.8 |
| Week 1 | 9.7 | 9.4 | 9.4 | 7.3 | 7.5 | 7.2 | 7.5 | 7.5 |
| Week 2 | 9.6 | 9.6 | 9.5 | 7.3 | 7.3 | 7.3 | 7.7 | 7.4 |
| Week 3 | 9.5 | 9.6 | 9.4 | 7.6 | 7.5 | 7.4 | 7.7 | 7.8 |

Table A-31: Analysis of variance on the TBA values for turkey bologna.

| Source | df | MS | F |
|---|---|---|---|
| Time (A) | 3 | 17.44 | 804.70** |
| Treatment (B) | 7 | 289.70 | 48.40** |
| A X B | 21 | 5.22 | 14.50** |
| Error | 224 | 0.36 | |
| Total | 255 | | |

**($p < 0.01$)

Table A-32: A comparison of treatment means for TBA values of turkey bologna using Dunnett's Test ($p < 0.01$)

| Time | Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| Week 0 | 5.26 | 5.26 | 5.58 | 1.90 | 1.35 | 2.19 | 1.55 | 1.56 |
| Week 1 | 9.35 | 8.16 | 8.07 | 1.80 | 1.43 | 2.06 | 1.50 | 1.37 |
| Week 2 | 7.89 | 8.52 | 7.34 | 1.67 | 1.57 | 2.12 | 1.48 | 1.72 |
| Week 3 | 8.00 | 8.51 | 7.80 | 2.25 | 1.44 | 1.84 | 1.48 | 1.43 |

Table A-33: Analysis of variance on the mean sensory scores of turkey ham color.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 117.67 | 2.39 |
| Treatments (B) | 5 | 16747.80 | 399.90** |
| Time (C) | 3 | 147.93 | 1.11 |
| A X B | 85 | 41.87 | 0.86 |
| A X C | 51 | 134.37 | 2.74 |
| B X C | 15 | 84.33 | 2.30 |
| A X B X C | 255 | 36.64 | 0.75 |
| Error | 432 | 49.04 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-34: A comparison of treatment means for turkey ham color using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 2.25 | 9.68 | 10.15 | 10.17 | 10.49 | 11.69 |

Table A-35: Analysis of variance on the mean sensory scores for turkey ham flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 18.43 | 1.30 |
| Treatment (B) | 5 | 245.46 | 19.07** |
| Time (C) | 3 | 35.92 | 1.86 |
| A X B | 85 | 12.87 | 0.91 |
| A X C | 51 | 19.29 | 1.36 |
| B X C | 15 | 8.34 | 1.13 |
| A X B X C | 255 | 7.36 | 0.52 |
| Error | 432 | 14.14 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-36: Analysis of variance on the mean sensory scores for turkey ham odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 425.63 | 3.88 |
| Treatment (B) | 5 | 4527.22 | 63.21** |
| Time (C) | 3 | 199.53 | 0.61 |
| A X B | 85 | 71.61 | 0.65 |
| A X C | 51 | 329.63 | 3.00 |
| B X C | 15 | 68.25 | 0.95 |
| A X B X C | 255 | 71.25 | 0.65 |
| Error | 432 | 109.79 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-37: A comparison of treatment means for turkey ham flavor using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 6.46 | 6.44 | 8.10 | 8.78 | 8.61 | 9.65 |

Table A-38: A comparison of treatment means for turkey ham odor using Dunnett's Test ($p < 0.01$)

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 4.47 | 8.34 | 7.99 | 7.54 | 6.87 | 9.78 |

Table A-39: Analysis of variance on the mean sensory scores for turkey ham off-flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 181.18 | 1.82 |
| Treatment (B) | 5 | 6524.63 | 111.11** |
| Time (C) | 3 | 635.82 | 2.28 |
| A X B | 85 | 58.72 | 0.59 |
| A X C | 51 | 278.79 | 2.79 |
| B X C | 15 | 44.10 | 0.64 |
| A X B X C | 255 | 69.07 | 0.69 |
| Error | 432 | 99.85 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-40: Analysis of variance on the mean sensory scores for turkey ham off-odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 250.11 | 2.66 |
| Treatment (B) | 5 | 3746.56 | 67.53** |
| Time (C) | 3 | 210.96 | 0.74 |
| A X B | 85 | 55.48 | 0.59 |
| A X C | 51 | 285.17 | 3.03 |
| B X C | 15 | 58.68 | 0.92 |
| A X B X C | 255 | 63.92 | 0.68 |
| Error | 432 | 93.94 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-41: A comparison of treatment means for turkey ham off-flavor using Dunnett's Test ($p < 0.01$).

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 7.13 | 3.76 | 2.28 | 2.08 | 2.46 | 1.14 |

Table A-42: A comparison of treatment means for turkey ham off-flavor using Dunnett's Test ($p < 0.01$).

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 5.93 | 2.55 | 2.21 | 2.40 | 2.36 | 1.24 |

Table A-43: Analysis of variance on the mean sensory scores for overall acceptability of turkey ham.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 17 | 16.45 | 2.26 |
| Treatment (B) | 5 | 1285.30 | 153.56** |
| Time (C) | 3 | 32.16 | 1.94 |
| A X B | 85 | 8.37 | 1.15 |
| A X C | 51 | 16.57 | 2.27 |
| B X C | 15 | 4.48 | 0.61 |
| A X B X C | 255 | 7.41 | 1.02 |
| Error | 432 | 7.29 | |
| Total | 863 | | |

**($p < 0.01$)

Table A-44: A comparison of treatment means for overall acceptability of turkey ham using Dunnett's Test ($p < 0.01$).

| Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|
| 00 | 20 | 40 | 60 | 100 | 156 |
| 2.68 | 7.83 | 9.19 | 9.59 | 9.45 | 11.22 |

Table A-45: Analysis of variance on Hunter Color L, $a_L$ and $b_L$ values for turkey ham.

| Source | df | MS | F |
|---|---|---|---|
| L Values | | | |
| Time (A) | 3 | 0.84 | 4.00** |
| Treatment (B) | 5 | 6.31 | 30.04** |
| A X B | 15 | 2.43 | 11.57** |
| Error | 72 | 0.21 | |
| Total | 95 | | |
| $a_L$ Values | | | |
| Time (A) | 3 | 0.32 | 8.95** |
| Treatment (B) | 5 | 63.35 | 171.20** |
| A X B | 15 | 1.10 | 2.97** |
| Error | 72 | 0.37 | |
| Total | 95 | | |
| $b_L$ Values | | | |
| Time (A) | 3 | 0.98 | 12.25** |
| Treatment (B) | 5 | 14.12 | 176.50** |
| A X B | 15 | 0.15 | 8.80** |
| Error | 72 | 0.08 | |
| Total | 95 | | |

**($p < 0.01$)

Table A-46: A comparison of treatment means for Hunter L, $a_L$ and $b_L$ values of turkey ham using Dunnett's Test ($p < 0.01$).

| Time | Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|---|
| | 00 | 20 | 40 | 60 | 100 | 156 |
| **L Values** | | | | | | |
| Week 0 | 43.5 | 43.1 | 43.2 | 42.6 | 43.3 | 42.4 |
| Week 1 | 43.5 | 42.4 | 43.3 | 43.4 | 42.4 | 42.6 |
| Week 2 | 46.2 | 42.3 | 42.9 | 43.1 | 43.2 | 42.3 |
| Week 3 | 44.3 | 42.8 | 43.5 | 42.2 | 42.5 | 44.3 |
| **$a_L$ Values** | | | | | | |
| Week 0 | 6.4 | 10.1 | 10.4 | 10.9 | 10.5 | 10.6 |
| Week 1 | 5.5 | 11.2 | 10.8 | 10.5 | 11.1 | 10.6 |
| Week 2 | 5.0 | 10.7 | 10.9 | 11.3 | 11.2 | 11.3 |
| Week 3 | 6.7 | 9.8 | 10.4 | 11.4 | 10.9 | 11.2 |
| **$b_L$ Values** | | | | | | |
| Week 0 | 10.1 | 8.1 | 7.9 | 7.9 | 8.0 | 8.0 |
| Week 1 | 9.7 | 7.1 | 7.8 | 7.8 | 7.4 | 7.6 |
| Week 2 | 10.6 | 7.9 | 7.9 | 7.7 | 7.9 | 7.9 |
| Week 3 | 10.0 | 7.9 | 7.9 | 7.8 | 7.8 | 7.8 |

Table A-47: Analysis of variance on the TBA values for turkey ham.

| Source | df | MS | F |
|---|---|---|---|
| Time (A) | 3 | 2.70 | 38.57** |
| Treatment (B) | 5 | 54.08 | 772.60** |
| A X B | 15 | 0.96 | 13.71** |
| Error | 168 | 0.07 | |
| Total | 191 | | |

**($p < 0.01$)

Table A-48: A comparison of treatment means for TBA values of turkey ham using Dunnett's Test ($p < 0.01$).

| Time | Treatment (mg/kg nitrite) | | | | | |
|---|---|---|---|---|---|---|
| | 00 | 20 | 40 | 60 | 100 | 156 |
| Week 0 | 3.00 | 0.47 | 0.22 | 0.11 | 0.18 | 0.25 |
| Week 1 | 3.20 | 0.75 | 0.34 | 0.44 | 0.21 | 0.19 |
| Week 2 | 2.90 | 0.49 | 0.26 | 0.18 | 0.19 | 0.12 |
| Week 3 | 4.90 | 0.78 | 0.48 | 0.43 | 0.36 | 0.28 |

Table A-49: Analysis of variance on the mean sensory scores of turkey ham color.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 7.94 | 2.15 |
| Treatment (B) | 7 | 868.92 | 235.50** |
| Time (C) | 3 | 7.25 | 1.69 |
| A X B | 63 | 3.50 | 0.95 |
| A X C | 27 | 4.30 | 1.17 |
| B X C | 21 | 4.90 | 1.33 |
| A X B X C | 189 | 3.69 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-50: A comparison of treatment means for turkey ham color using Dunnett's Test ($p < 0.01$).

| Trestment | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 2.54 | 2.20 | 2.52 | 11.40 | 11.34 | 11.12 | 11.53 | 11.71 |

Table A-51: Analysis of variance on the mean sensory scores of turkey ham flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 13.78 | 1.62 |
| Treatment (B) | 7 | 461.72 | 48.35** |
| Time (C) | 3 | 3.33 | 0.17 |
| A X B | 63 | 9.55 | 1.12 |
| A X C | 27 | 20.21 | 2.38 |
| B X C | 21 | 11.07 | 1.30 |
| A X B X C | 189 | 8.49 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-52: Analysis of variance on the mean sensory scores for turkey ham odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 4.44 | 0.46 |
| Treatments (B) | 7 | 432.04 | 52.40** |
| Time (C) | 3 | 0.55 | 0.03 |
| A X B | 63 | 8.25 | 0.85 |
| A X C | 27 | 21.03 | 2.16 |
| B X C | 21 | 12.40 | 1.27 |
| A X B X C | 189 | 9.75 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-53: A comparison of treatment means for turkey ham flavor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 4.27 | 2.79 | 2.32 | 10.05 | 9.71 | 8.78 | 8.37 | 10.48 |

Table A-54: A comparison of treatment means for turkey ham odor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 3.70 | 3.34 | 3.35 | 10.29 | 9.20 | 10.16 | 9.16 | 9.98 |

Table A-55: Analysis of variance on the mean sensory scores for turkey ham off-flavor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 21.34 | 2.09 |
| Treatments (B) | 7 | 416.61 | 41.54** |
| Time (C) | 3 | 14.58 | 0.65 |
| A X B | 63 | 10.03 | 0.98 |
| A X C | 27 | 22.54 | 2.21 |
| B X C | 21 | 16.74 | 1.64 |
| A X B X C | 189 | 10.22 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-56: Analysis of variance on the mean sensory scores for turkey ham off-odor.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 7.70 | 0.69 |
| Treatments (B) | 7 | 431.47 | 56.20** |
| Time (C) | 3 | 28.85 | 2.53 |
| A X B | 63 | 7.68 | 0.69 |
| A X C | 27 | 11.41 | 1.04 |
| B X C | 21 | 16.08 | 1.46 |
| A X B X C | 189 | 11.02 | |
| Total | 319 | | |

**($p < 0.01$)

Table A-57: A comparison of treatment means for turkey ham off-flavor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 7.79 | 8.44 | 9.02 | 1.65 | 2.09 | 3.93 | 2.82 | 1.46 |

Table A-58: A comparison of treatment means for turkey ham off-odor using Dunnett's Test ($p < 0.01$).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 7.60 | 8.40 | 8.02 | 1.41 | 1.78 | 1.96 | 1.64 | 1.60 |

Table A-59: Analysis of variance on the mean sensory scores for turkey ham overall acceptability.

| Source | df | MS | F |
|---|---|---|---|
| Evaluators (A) | 9 | 25.76 | 3.56 |
| Treatment (B) | 7 | 601.45 | 72.29** |
| Time (C) | 3 | 10.90 | 0.80 |
| A X B | 63 | 8.32 | 1.15 |
| A X C | 27 | 13.62 | 1.88 |
| B X C | 21 | 9.69 | 1.34 |
| A X B X C | 189 | 7.24 | |
| Total | 319 | | |

**(p 0.01)

Table A-60: A comparison of treatment means for turkey ham overall acceptability using Dunnett's Test (p 0.01).

| Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|
| 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| 2.94 | 2.66 | 2.15 | 10.85 | 9.84 | 8.34 | 9.32 | 11.06 |

Table A-61: Analysis of variance on Hunter Color L, $a_L$ and $b_L$ values for turkey ham.

| Source | df | MS | F |
|---|---|---|---|
| L Values | | | |
| Time (A) | 3 | 3.32 | 27.68** |
| Treatment (B) | 7 | 25.53 | 212.75** |
| A X B | 21 | 2.03 | 16.92** |
| Error | 96 | 0.13 | |
| Total | 127 | | |
| $a_L$ Values | | | |
| Time (A) | 3 | 13.72 | 274.40** |
| Treatment (B) | 7 | 50.91 | 1018.20** |
| A X B | 21 | 17.71 | 354.20** |
| Error | 96 | 0.05 | |
| Total | 127 | | |
| $b_L$ Values | | | |
| Time (A) | 3 | 8.89 | 296.33** |
| Treatment (B) | 7 | 1.95 | 65.00** |
| A X B | 21 | 19.27 | 642.33** |
| Error | 96 | 0.03 | |
| Total | 127 | | |

**($p < 0.01$)

Table A-62: A comparison of treatment means of Hunter L, $a_L$ and $b_L$ turkey ham using Dunnett's Test ($p < 0.01$).

| Time | Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| **L Values** | | | | | | | | |
| Week 0 | 43.4 | 42.4 | 42.1 | 41.4 | 39.8 | 40.8 | 40.3 | 40.7 |
| Week 1 | 42.6 | 42.4 | 42.2 | 41.3 | 40.1 | 40.7 | 39.1 | 40.5 |
| Week 2 | 42.2 | 41.7 | 43.1 | 40.5 | 40.0 | 39.2 | 38.4 | 39.7 |
| Week 3 | 42.6 | 42.9 | 41.8 | 41.6 | 38.9 | 39.8 | 41.6 | 39.4 |
| **$a_L$ Values** | | | | | | | | |
| Week 0 | 5.9 | 6.0 | 5.3 | 11.2 | 11.5 | 11.0 | 11.5 | 11.5 |
| Week 1 | 5.5 | 5.8 | 5.7 | 11.2 | 11.5 | 11.3 | 11.4 | 11.4 |
| Week 2 | 6.4 | 5.8 | 6.2 | 11.6 | 11.8 | 11.9 | 11.7 | 11.9 |
| Week 3 | 6.2 | 6.6 | 6.4 | 11.4 | 11.9 | 11.5 | 11.3 | 11.7 |
| **$b_L$ Values** | | | | | | | | |
| Week 0 | 9.7 | 10.0 | 10.2 | 7.4 | 7.1 | 7.1 | 7.2 | 7.2 |
| Week 1 | 10.2 | 9.9 | 9.6 | 7.3 | 7.2 | 7.3 | 7.3 | 7.4 |
| Week 2 | 9.9 | 9.8 | 10.2 | 7.1 | 7.0 | 7.0 | 6.7 | 7.0 |
| Week 3 | 10.0 | 9.9 | 9.6 | 7.3 | 6.9 | 7.0 | 7.5 | 7.4 |

Table A-63: Analysis of variance on the TBA values for turkey ham.

| Source | df | MS | F |
|---|---|---|---|
| Time (A) | 3 | 2.96 | 24.67** |
| Treatment (B) | 7 | 75.25 | 627.08** |
| A X B | 21 | 2.76 | 23.00** |
| Error | 224 | 0.12 | |
| Total | 255 | | |

**(p< 0.01)

Table A-64: A comparison of treatment means for TBA values of turkey ham using Dunnett's Test (p < 0.01).

| Time | Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 00 | 02 | 03 | 40 | 60 | 42 | 62 | 156 |
| Week 0 | 3.50 | 3.50 | 2.60 | 0.54 | 0.61 | 0.64 | 0.43 | 0.52 |
| Week 1 | 3.30 | 4.60 | 3.20 | 0.53 | 0.37 | 0.40 | 0.28 | 0.33 |
| Week 2 | 2.70 | 3.60 | 2.00 | 0.64 | 0.49 | 0.78 | 0.59 | 0.71 |
| Week 3 | 3.70 | 3.70 | 5.40 | 0.86 | 0.29 | 0.49 | 0.64 | 0.46 |