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PROCESSING OF WILTSHIRE BACON WITH
 α -TOCOPHEROL-COATED SALTS

By
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ABSTRACT

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By

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The efficacy of α -tocopherol-coated salts as curing adjuncts in immersion-cured and dry-cured Wiltshire bacon was determined. The quantity of N-nitrosopyrrolidine and N-nitrosodimethylamine formed in cooked rashers was determined and the sensory and microbiological qualities of the bacon were evaluated. The addition of α -tocopherol-coated salts to Wiltshire bacon markedly reduced the quantity of N-nitrosamine formed in both immersion-cured and dry-cured bacon samples. N-Nitrosamine levels in bacon decreased with time of storage, and this decrease was correlated to the decrease in residual nitrite concentration. Sensory evaluation of the bacon samples indicated that the addition of α -tocopherol did not influence the organoleptic qualities of the bacon. In general, Wiltshire bacon cured with and without α -tocopherol-coated salts were similar in patterns of microbial growth.

Dedicated to
my fiance

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INTRODUCTION

Wiltshire bacon is the form of bacon most widely sold in the United Kingdom, and can be purchased in the form of collar and back bacon which are produced from separate areas on the pig carcass. Collar bacon is prepared from the area forward from the middle of the shoulder pocket to the jowl, while back bacon is prepared from the elbow joint to the end of the last rib. Wiltshire bacon typically has nitrate, nitrite, and salt levels of 500 mg/kg, 100 mg/kg, and 4 to 5%, respectively. More recently, it has been shown that the exclusion of nitrate from the curing ingredients is not deleterious to the resultant product (Taylor and Shaw, 1975).

Two predominant methods for processing Wiltshire bacon currently exist. The traditional immersion method of curing typically requires 10 days for the manufacture of the finished product. A more modern style of Wiltshire bacon processing involves needle injection of brine followed by dry salt curing and results in a finished product in as little as 5 days (Taylor et al., 1980).

Fried bacon as well as the cooked-out fat typically have been a major source of N-nitrosamines (Gray, 1981). N-Nitrosamine levels in Wiltshire bacon have been reported to be as much as 15.6 ug/kg and 6.7 ug/kg for N-nitrosopyrrolidine (NPYR) and N-nitrosodimethylamine (NDMA), respectively (Mottram et al., 1977). The inclusion of anti-N-nitrosamine agents in bacon such as α -tocopherol has been

effective in inhibiting the formation of N-nitrosamines in bacon produced from pork bellies by 60 - 90% (Skrypec et al., 1985, Bernthal et al., 1986).

From a microbiological standpoint, Wiltshire bacon which has been dry cured has been reported to contain significantly lower ($P < 0.01$) counts of total viable bacteria in comparison to bacon which had been cured by the traditional immersion method (Taylor et al., 1980). There also seemed to be a tendency for dry-cured Wiltshire bacon to initially have lower counts of lactic acid bacteria on collar and back bacon than their immersion-cured counterparts. The increase in lactic acid bacteria counts observed in the dry cured back bacon during storage can be attributed to the lower concentrations of curing salts found in that region of the carcass.

The major focus of this study is to evaluate the inclusion of α -tocopherol as a curing adjunct in the processing of Wiltshire bacon by both immersion and dry-cure processes. The effect of α -tocopherol on the inhibition of N-nitrosamine formation during the frying of bacon will be investigated, as will its effect on the microbiological quality of the finished bacon. Additionally, organoleptic evaluation of Wiltshire bacon prepared with and without α -tocopherol-coated salts will be addressed.

LITERATURE REVIEW

The curing of bacon is as much an art as it is a science. Bacon taste, appearance, desirability and its preservation are some of the factors which can be controlled in part by the curing method practiced (Kramlich et al., 1973). In addition, it is important to choose a method of processing which will result in high yields, a rapid turnover rate, reproducibility and uniformity, and finally a processing procedure that is the least labor intensive. A successful bacon processor will satisfy all or most of these criteria.

Processing of Bacon

Processing by dry salt. The oldest known method of curing meat or fish is by rubbing salt on the surface of the meat. The curing of pork was traditionally accomplished by rubbing the surface with a mixture of salt and nitrate (saltpeter) (Taylor and Shaw, 1975). Juices in the meat dissolve the salt mixture and the cure is distributed throughout the tissue by means of diffusion. Although dry salting meat extends the shelf life of the product, meats treated only with salt tend to be dry, harsh and overly salty (Kramlich et al., 1973). Dry salted meats also tend to have a dark undesirable color (Kramlich et al., 1973).

A more modern style of dry salting utilizes sugar and nitrite/nitrate in addition to salt. Nitrite decreases microorganisms present in the meat, contributes to the characteristic flavor of bacon,

and is responsible for color development of the bacon (Kerr et al., 1926). Thus, the use of nitrite as a curing ingredient eliminates some of the problems associated with the use of salt by itself.

Dry salting as a method of curing is safe and simple to perform. A relatively long period of time is required, however, for the diffusion of the cure into the meat. Approximately 7 days per inch of thickness is required for curing bellies (Kramlich et al., 1973). This is one of the major disadvantages of the dry curing technique.

Processing by needle injection. Needle injection (stitch pumping) of bellies can be accomplished by either a single or a multineedle injection apparatus (Kramlich et al., 1973). In general, the curing ingredients are dissolved in the brine and the brine is delivered through openings at the tips of needle(s) which have been plunged into the muscle tissue. Constituents of the brine include salt, sugar, nitrite and/or nitrate, and phosphates. Phosphates are added to aid in the retention of water, thereby increasing yields. Bellies are usually pumped to 110% of their green weight using a warm (approximately 65°C) curing solution (Kramlich et al., 1973). Pork bellies are very well suited to needle injection in that their thinness allows brine to diffuse rapidly throughout the tissue (Kramlich et al., 1973).

Reduced labor costs and better yields have increased the popularity of stitch pumped meats. It is estimated that 80% of bacon produced in the United States is processed in this manner (Kramlich et al., 1973). Some of the disadvantages of needle injection, however, include increased cooking shrinkage and decreased organoleptic acceptability.

Processing by pickle curing. Salt, sugar, nitrite and/or nitrate are dissolved in water and contained in a large vat to function as the pickle cure. Pickle curing, often called immersion curing, involves the submerging of meat in the brine for up to 2 weeks. Although curing practices differ in the length of time the meat is in the pickle and in the concentration of curing ingredients in the pickle (Cook and White, 1940), the overall result is a uniformly cured product that is milder in taste than the dry cured product (American Meat Institute, 1944; Kramlich et al., 1973). Combination curing is combining stitch pumping or artery pumping with either dry salting or immersion curing. By combining methods of curing, a more rapid rate of product output is achieved (Kramlich et al., 1973).

Thermal or hot processing. In an effort to decrease the curing time even further, a thermal cure technique has been developed (Kramlich et al., 1973). This method utilizes hot dry cures or hot pickles. The concept behind hot processing is that curing is accelerated at higher temperatures, thus decreasing the total curing time. Dry cure temperatures should be 48-50°F while immersion cure temperatures are higher (135-140°F) (Kramlich et al., 1973).

Another variation of hot processing is to begin the curing process on freshly slaughtered and cleaned carcasses before rigor mortis has occurred (Cuthbertson, 1982). The advantages of hot processing pre-rigor meat are many and include greater yields, more uniform lean color, more marketable meat, less drip in vacuum package, reduced processing times and lower labor costs (Cuthbertson, 1982). A major cost reductant is in refrigeration costs, both capital and running (Cuthbertson, 1982; Kastner, 1982). Stringent hygiene and temperature

controls are necessary to insure a quality meat product processed by the hot processing technique (Cuthbertson, 1982).

Although there are several advantages to the hot processing of meat, many processors are reluctant to practice such a method. In the United States, the standards set by the United States Department of Agriculture (USDA) for beef carcasses are based on chilled meat (Cuthbertson, 1982). In Britain, classification and grading of meat is frequently based on warm sides but the processors are wary of the research findings that support hot processing (Cuthbertson, 1982). The British also express concern for microbial contamination of hot processed meat. Nevertheless, hot processing of meat of beef, pork and lamb origin is being researched extensively (Cuthbertson, 1982; Kastner, 1982).

Types of Wiltshire bacon. Wiltshire bacon is a common commodity in the United Kingdom and is sold sliced and sealed in a vacuum packaged bag. Wiltshire bacon is sold either smoked or unsmoked. The finished product contains 4 to 5% salt (Taylor et al., 1980) which is much more salty than bacon produced in the United States (Kramlich et al., 1973).

Wiltshire bacon consists of mainly three types of bacon. Collar and back bacon are sold in a sliced and vacuum packaged state. The hind legs, the gammons, are usually sold separately (Kramlich et al., 1973). Bacon prepared from the area forward from the middle of the shoulder pocket to the jowl is designated as collar bacon. Back bacon is taken from the elbow joint to the end of the last rib. As shown in Figure 1, bacon produced from these areas of the carcass will be meaty and contain very little fat. Back bacon, however, contains more fat

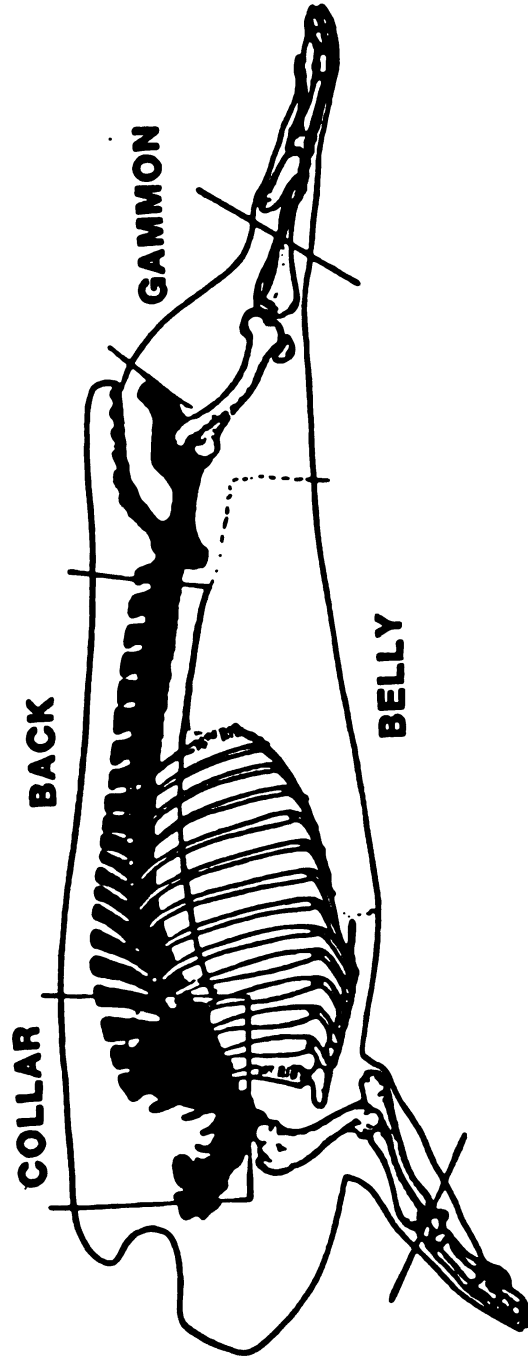


Figure 1. A Wiltshire bacon side.

than does collar bacon, but the fat is mainly restricted to the layer of backfat and rind. In contrast, bacon produced in the United States is prepared from the belly area and is relatively high in fat content, containing up to 42 - 63% adipose per total weight of the pork belly (Gray, 1976). Collar and back bacon is sliced to approximately 3 mm of thickness after curing. Sliced bacon is packaged under vacuum and stored at 4°C.

Wiltshire bacon curing levels. The British Bacon Curers' Federation Code of Practice recommends the inclusion of both nitrite and nitrate in brines used for Wiltshire curing (Taylor and Shaw, 1975). Nitrite levels of 1000 mg/kg and nitrate at 5000 mg/kg are dissolved in the brine and result in finished product levels of 100 mg/kg and 500 mg/kg, respectively. Traditionally, Wiltshire sides were processed using 2000-3000 mg/kg of nitrate in the cured meat, but has since been limited to 500 mg/kg (Taylor and Shaw, 1975). The reduction of nitrate was due to the potentially high source of nitrite which is known to cause health hazards (Statutory Instruments, 1971). Levels of sodium chloride in the brine are typically 26% wt/vol with a finished product level of approximately 4% (Taylor and Shaw, 1975).

Recently, researchers have become interested in decreasing or omitting nitrate in the curing ingredients (Taylor and Shaw, 1975; Taylor et al., 1976). Researchers have found bacon cured with brine containing 1000 mg/kg nitrite and no nitrate to be satisfactorily acceptable after storage for 5 weeks at 5°C or 2 weeks at 15°C (Taylor and Shaw, 1975).

Dressing procedure for Wiltshire bacon. Although many variations of the basic processing steps for Wiltshire bacon have been practiced

(Taylor et al., 1980), the dressing procedure for Wiltshire bacon sides has remained fairly constant. Most Wiltshire sides are produced from hogs weighing 68 - 90 kg live weight. After slaughter, the carcass is split in half lengthwise, the head is removed along with the backbone, aitchbone, tenderloin, skirt and ribs. The forelegs and hindlegs are trimmed at the knees and the hocks, respectively. After curing, the hind legs are often removed from the carcass and sold separately as gammons. The ideal Wiltshire side weighs 23 - 27 kg and has a backfat thickness between 3.2 - 3.8 cm after trimming (Kramlich et al., 1973).

History of Wiltshire curing. Up until the 1930's, Wiltshire bacon had been cured by rubbing dry salt and saltpeter (potassium nitrate) onto the surfaces of the meat (Taylor et al., 1980). Curing of the meat was accomplished by microbiological reduction of the nitrate to nitrite. This method of curing tended to be lengthy and laborious, thus immersion curing of Wiltshire sides soon became the established method of curing.

The oldest style of immersion curing Wiltshire sides involved hand injection of the side with a solution containing salt and nitrate, then submerging the side into a large vat containing a similar curing solution. The side would remain submerged for approximately 5 days, and then hung and allowed to mature for up to 2 weeks (Taylor et al., 1980). Curing time for Wiltshire sides has since been reduced to 7 to 10 days through the use of the multineedle injection format of curing (Taylor et al., 1976, 1980). This 7 day processing technique, coined the "conventional method," is currently the most popular form of curing Wiltshire bacon in the United Kingdom (Taylor et al., 1980).

Immersion curing of Wiltshire bacon. Procedures for processing Wiltshire bacon are outlined in Figure 2 and summarized as follows: After slaughter, the carcass is held overnight at 4°C, then dressed for Wiltshire bacon as previously described. Sides are then pumped with brine using a multineedle machine to an average of 10% increase in weight. Following brine injection, the sides are then immersed in a pickle. After 3 days in the brine, the sides are removed, hung by the hind leg and allowed to mature for 7 days (Taylor et al., 1980).

Modern dry salt curing of Wiltshire bacon. A reduction in total processing time of Wiltshire sides can be accomplished by a combination curing method of multineedle injection, followed by dry salting (Taylor et al., 1980). This modern dry-salting process allows the final product to be marketed after only 5 days. An outline of the dry salting method as described by Taylor et al., 1980 is illustrated in Figure 2. Advantages of the modern dry-salting process include lower initial microbial load, decreased labor and decreased production space (Taylor et al., 1980). Taylor et al. (1980) reported that bacon processed in this manner was similar in eating quality, yields and appearance to that processed by the conventional technique. They also found that dry salted, vacuum packaged collar bacon stored slightly better than its back bacon counterpart. They attributed this to a lower curing salt concentration in the back bacon compared to the collar bacon. Additionally, Taylor et al. (1980) found that back bacon in immersion cured sides contained slightly higher levels of curing salts than collars from the same side. This phenomenon was also observed by Patterson et al. (1976) who found that a gradient of curing salts in the bacon with abnormally high concentrations of nitrite in

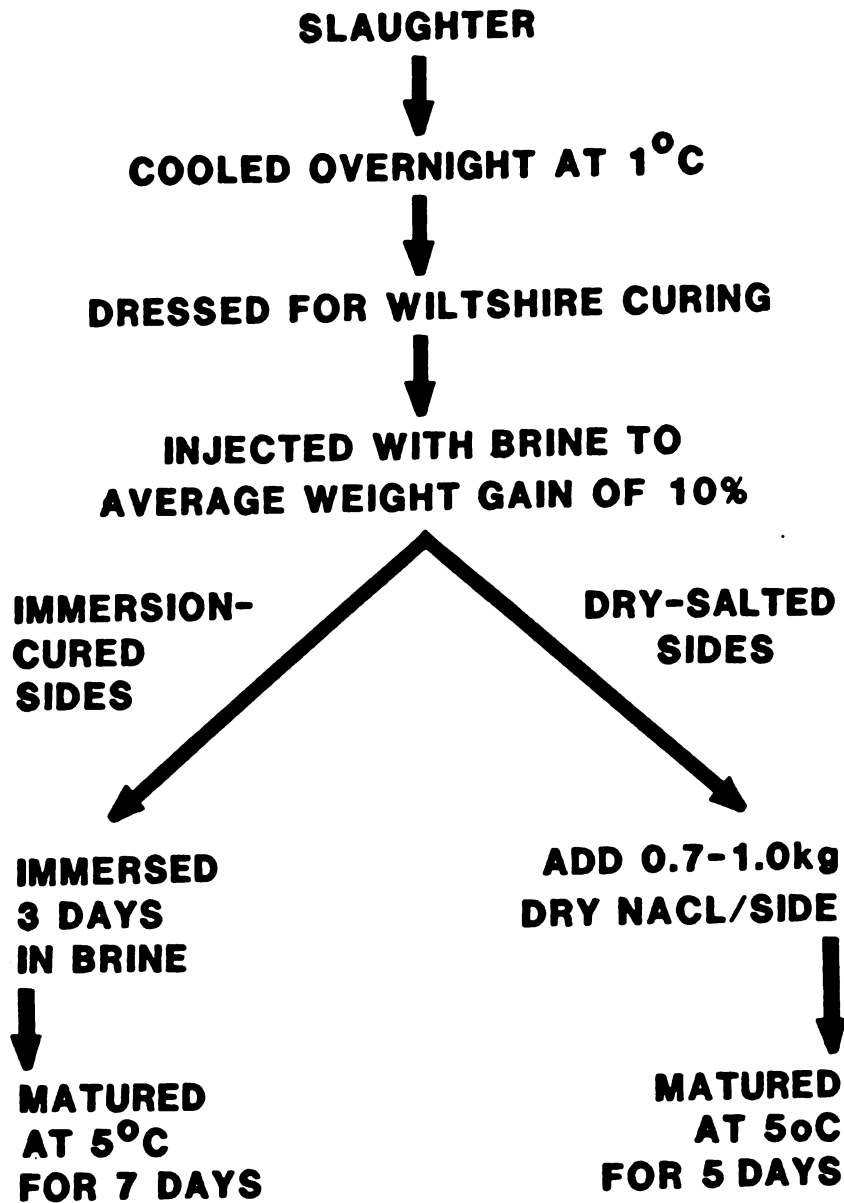


Figure 2. General processing procedure for Wiltshire bacon (Taylor et al., 1980).

the outer layers of bacon sides. Bacon processors practicing the modern dry-curing method should realize the unevenness of curing salt uptake and compensate by increasing the levels of cure applied to the back bacon area of the carcass (Taylor et al., 1980). Such a system would have the advantage of a more controlled distribution of curing salt uptake and would result in a more consistent product (Taylor et al., 1980).

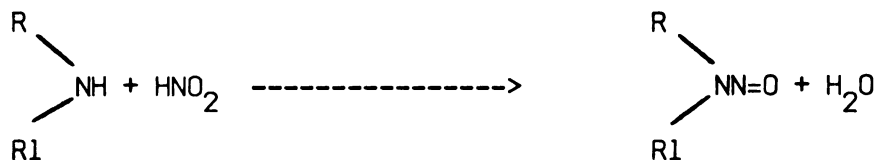
N-Nitrosamines in Bacon

N-Nitrosamines in food products. Ender et al. (1964) identified a compound from toxic herring meal that has since been the cause of considerable research. The compound isolated was N-nitrosodimethylamine (NDMA). They theorized that its formation stemmed from the reaction between sodium nitrite and dimethylamine. Subsequent research has shown that N-nitrosamines are formed primarily from the reaction between nitrite and secondary amines (Mirvish, 1970; Gray, 1981).

N-Nitrosamines have been shown to be carcinogenic in laboratory animals by a number of researchers (Magee and Barnes, 1956; Crosby and Sawyer, 1976). Moreover, some N-nitroso compounds are mutagenic, teratogenic and embryopathic (Gray, 1981; Scanlan and Reyes, 1985). Since amines are found in nearly all foods, and nitrosating agents also commonly occur in foods, the presence of N-nitrosamines in the human food supply was questioned. Among the foods that have been found to contain N-nitrosamines are cured meat products, beer, dry malt, non-fat dry milk and soy protein foods (Gray and Randall, 1979; Havery and Fazio, 1985).

N-Nitrosamines in cured meat products. Cured meat products, bacon in particular, have been extensively studied since the early 1970's (Gray, 1981). NDMA and N-nitrosopyrrolidine (NPYR) have been consistently isolated from both cooked bacon and the cooked-out fat (Table 1). Additionally, N-nitrosamines have been found in the fumes that have been produced during the frying of bacon (Table 2). Raw bacon does not contain NPYR, whereas cooked bacon contains considerable quantities of NPYR and lesser quantities of NDMA.

Formation of N-nitrosamines. N-Nitrosamines are formed from the reaction of nitrous acid and secondary amines as shown below:



Detailed reviews of the chemistry of the reactions of N-nitrosamine formation are available (Crosby and Sawyer, 1976; Gray and Randall, 1979; Gray, 1981). Of all the N-nitrosamines, NPYR is the most widely studied. Presently, it is believed that NPYR is formed by nitrosation of proline, followed by decarboxylation of N-nitrosoproline during the frying process (Bharucha et al., 1979; Lee et al., 1983).

Factors affecting N-nitrosamine formation in bacon. Factors which affect the formation of N-nitrosamines in fried bacon have been reviewed by several researchers including Gray (1976); Gray and Randall (1979); and Sen (1980). These factors include residual nitrite, processing procedures, cooking methods, frying temperature and time, the fat to lean ratio and fatty acid composition, ascorbate concentration, and N-nitrosamine inhibitors.

Table 1. N-Nitrosamine levels (ug/kg) in fried bacon.^a

Investigators	NPYR		NDMA	
	(ug/kg)			
	Bacon	Cooked-out fat	Bacon	Cooked-out fat
Crosby et al. (1972)	tr-40	--	tr	--
Sen et al. (1973)	4-25	--	2-30	--
Fiddler et al. (1974)	2-28	6-24	--	--
Pensabene et al. (1974)	11-38	16-39	--	--
Gray et al. (1977)	tr-23	tr-41	--	--
Pensabene et al. (1979a)	2-45	5-55	2-9	2-34
Sen et al. (1979)	2-22	15-34	tr-17	3-12
Pensabene et al. (1980)	2-6	11-34	--	--

^aAdapted from Gray (1981).

Table 2. Percentage of total N-nitrosamines produced during the frying of bacon which are present in the fumes.^a

Investigators	N-Nitrosamine (%)		
	NPYR	NDMA	Sample
Gough et al. (1976)	60-95	75-100	bacon
Hwang and Rosen (1976)	14-37	----	bacon
Warthesen et al. (1976)	20-40	----	pork belly ^b
Sen et al. (1976)	28-82	28-92	bacon
Gray and Collins (1977)	27-49	----	pork belly ^b
Mottram et al. (1977)	57-75	73-80	bacon
Gray et al. (1978)	----	56-80	pork belly ^b
Bharucha et al. (1979)	up to 32	up to 62	bacon

^aAdapted from Gray (1981).

^bContained added nitrite.

Residual nitrite. The source of nitrous acid is nitrite, which is an integral ingredient of the cure. Nitrite may also be formed as the result of microbial reduction of nitrate (Gray, 1981), another cure additive which is frequently used in the processing of Wiltshire bacon sides.

Since nitrite is so basically involved in the production of N-nitrosamines, it is not surprising that nitrite concentration affects the levels at which N-nitrosamines are formed. Sen et al. (1974) suggested that formation of N-nitrosamines was dependent on the initial level of nitrite and not the final (residual nitrite) concentration. However, subsequent research has indicated that the residual nitrite concentration is responsible for the levels of volatile N-nitrosamines which are produced (Dudley, 1979; Sebranek, 1979). More interestingly, less than 50% of the added nitrite in a meat system can be accounted for after the completion of processing (Cassens et al., 1974).

Realizing the potential health risks associated with high levels of nitrites, it was recommended that the levels of in-going nitrite be reduced from 156 to 120 mg/kg (Federal Register, 1975). The nitrate concentration in Wiltshire bacon has been reduced from 2000-3000 to 500 mg/kg (Taylor and Shaw 1975). Canadian bacon processors have seen a similar decrease in allowable nitrite levels, 150 mg/kg, down from 200 mg/kg (Gray, 1976) (Table 3).

Processing procedures. It has been shown that the method of curing bacon affects the concentration of N-nitrosamines isolated after frying. Pensabene et al. (1979) reported that over time, there was an increase in the free proline detected in dry cured bacon and that this

Table 3. Maximum allowable nitrite and nitrate concentrations in various bacon types.

Bacon Type	Nitrite (mg/kg)	Nitrate (mg/kg)
Belly bacon (USA) ^a	120 ¹	---
Canadian bacon ^b	150	---
Wiltshire bacon (UK) ^c	100	500

^aFederal Register, 1975, 40, 52614

^bCanadian Food and Drug Regulations, amendment April 1975 (B. 16. 100, Items P1 and P2, Table X1, Part 1).

^cBritish Bacon Curers' Federation Code of Practice (Statutory Instruments, 1971).

¹Mandatory level of nitrite to be used in conjunction with 550 mg/kg sodium ascorbate (Federal Register, 1975).

proline correlated with high levels of N-nitrosamines found in the fried bacon.

Although Pensabene et al. (1979) concluded from their study that free proline may play a significant role in the formation of NPYR in dry cured products, it is also important to note that the NPYR concentration in fried dry cured bacon is higher than that formed in other types of bacon (Table 4). High levels of NPYR in dry cured bacon have also been reported by the Nitrite Safety Council (1980).

Cooking methods. Pensabene et al. (1974) reported that N-nitrosamines were formed in bacon at much lower (almost negligible) quantities when microwaved compared to bacon which was fried, baked or broiled. They suggested that the small concentrations formed in

Table 4. N-Nitrosamine concentrations in cured meat products.^a

Sample type	NPRO ^{b,c} (ug/kg)	NPYR ^d (ug/kg)
Bacon	N.D. ^e	5-63
Dry cured bacon	106	39-89
Canadian bacon	N.D.	N.D.
Pork side meat	86-411	19-149

^aAdapted from Pensabene et al. (1979)

^bMinimum level of detection 10 ug/kg

^cUncooked samples

^dAfter frying samples

^eN.D. = None detected

microwaved bacon may be due to the short exposure time of the meat to heat. In a later study, grilled bacon was found to contain less N-nitrosamines than fried bacon (Bharucha et al., 1979). It was concluded that during grilling, the high temperatures that are achieved during pan-frying of bacon are not reached when bacon is grilled. This is due to the fact that the cooked-out fat runs out of the heated area, and therefore, the higher temperatures realized in pan frying are never reached.

Frying temperature and time. The formation of NPYR as a result of various frying treatments was researched by Pensabene et al. (1974). They found that bacon fried at 99°C for 105 minutes formed no NPYR, while an identical sample fried at 204°C for 4 minutes produced 17

ug/kg of NPYR. Both of these bacon samples were fried to medium well-done.

There appears to be a critical time when the maximum amount of N-nitrosamines are formed during frying. Combining all three fractions (rashers, cooked-out fat and condensed vapor), Hotchkiss and Vecchio (1985) reported that the maximum amounts of NDMA and NPYR were formed after frying for three minutes per side in a preheated skillet (171°C). The values obtained were 27.5 and 58.0 ug/kg of raw bacon for NDMA and NPYR, respectively. Longer periods of frying decreased the total amounts of NDMA and NPYR that were recovered. This was probably due to the thermal decomposition of the N-nitrosamines (Hotchkiss and Vecchio, 1985). Thermal decomposition of NPYR in heated bacon was also observed by Nakamura et al. (1976). Bharucha et al. (1979) found that maximum levels of NPYR are produced after bacon is fried or grilled for 12 minutes using a non-preheated frying pan (360°F). Initially, very small quantities of NPYR were observed in the cooked-out fat, then the concentration of NPYR began to increase at 4 minutes of frying, reaching a maximum at 12 minutes. After 12 minutes, the NPYR concentration began to decline. Two explanations for the initial low formation of N-nitrosamines were offered: (1) that the N-nitrosamines are actually formed at about 100°C but, being steam-volatile, are removed with the expelled water, or (2) that the nitrosation occurs at temperatures greater than 100 after the major portion of the water is removed and therefore occurs in the fat phase (Bharucha et al., 1979).

Fat to lean ratio and fatty acid composition. Bacon with a high fat to lean ratio has been found to yield higher concentrations of NPYR than bacon having low ratios (Pensabene et al., 1979). Fiddler et al.

(1974) resolved that NPYR was derived from the adipose tissue after extensively studying both the lean and fat components of the bacon before and after frying. They found that NPYR was detected only in the fat component after frying and was not found in either the unfried fat or the unfried or fried lean component. It was concluded that the precursor for NPYR formation must arise in the adipose tissue of bacon.

Skrypec et al. (1985) concluded that belly bacon from pigs fed a corn oil-enriched diet contained significantly greater levels of NPYR than bacon produced from pigs fed a control diet. They also observed a decrease in NPYR production in bacon produced from pigs fed a coconut oil-enriched diet. Since corn oil and coconut oil contained highly unsaturated and highly saturated fatty acids, respectively, it was concluded that the degree of saturation of fatty acids in the adipose tissue affects the levels of NPYR produced in bacon (Skrypec et al., 1985).

Ascorbate concentration. In an effort to decrease the levels of N-nitrosamines produced during the frying of bacon, it was recommended that sodium ascorbate be included in the brine at an ingoing concentration of 550 mg/kg (Federal Register, 1975). Simultaneous to the addition of sodium ascorbate in the brine, there was a decrease in the nitrite permitted to be added to bacon in processing (i.e., from 156 mg/kg to 120 mg/kg (Federal Register, 1975).

It has been shown that sodium ascorbate decreases NPYR production in fried bacon (Greenberg, 1973). Sodium ascorbate and sodium erythorbate have also been found to inhibit N-nitrosamine formation in frankfurters (Fiddler et al., 1973). Wiltshire bacon spiked with dimethylamine and cured with an ascorbate-containing brine contained

lower quantities of NDMA than control (non ascorbate-cured) bacon (Mottram et al., 1975).

Mottram and Patterson (1977) found that in a model system, sodium ascorbate increased nitrosation by 5 to 25 times. The addition of ascorbyl palmitate to the model system, however, generally reduced N-nitrosamine formation. The lipophilic character of ascorbyl palmitate may be one of the major factors in its ability to decrease N-nitrosamine formation.

N-Nitrosamine inhibitors. Recent studies have indicated that an effective anti-nitrosamine agent for bacon must be (1) able to trap NO radicals; (2) lipophilic; (3) non-steam volatile; and (4) heat stable up to 174°C (maximum frying temperature) (Bharucha et al., 1979). Several compounds having these characteristics have been studied for their effectiveness as anti-N-nitrosamine agents. Ascorbyl palmitate has been studied by Sen et al. (1976), while long chain acetals of ascorbic acid have been researched by Bharucha et al. (1980). Another compound, α -tocopherol, has been shown to be effective against N-nitrosamine formation by several researchers (Fiddler et al., 1978; Gray et al., 1982; Reddy et al., 1982).

Fiddler et al. (1978) found that the inclusion of α -tocopherol in the curing brine, alone or in combination with sodium ascorbate, inhibited the formation of NPYR and NDMA in bacon more effectively than did the addition of ascorbate alone. The inclusion of 500 mg/kg α -tocopherol has been found to inhibit the formation of N-nitrosamines in fried bacon by up to 80% (Fiddler et al., 1978). Gray et al. (1982) reported that α -tocopherol, when used in the processing of brine-cured bacon, inhibited N-nitrosamine formation by 87 and 97%, respectively,

in fried bacon and the cooked-out fat. N-Nitrosamines present in the cooking vapors have also been reduced in bacon fried in fat containing α -tocopherol (Walters et al., 1976). Mergens and Newmark (1979) reported a four-fold suppression of N-nitrosamines in the cooking vapors when bacon was cured in the presence of 500 mg/kg α -tocopherol.

α -Tocopherol-coated salts of high surface area, when used in the processing of dry-cured bacon, inhibited NPYR formation by up to 96% (Gray et al., 1982; Reddy et al., 1982). Results from these studies indicate that the optimum level of ingoing α -tocopherol is 500 mg/kg, as higher or lower levels of α -tocopherol resulted in lower inhibition levels. In general, the effect of α -tocopherol on the inhibition of N-nitrosamine formation was much greater for NPYR than for NDMA for either dry cured or brine cured bacon (Fiddler et al., 1978; Gray et al., 1982; Reddy et al., 1982).

Dispersion of α -tocopherol throughout the bacon is important for optimum inhibition of N-nitrosamines. Pensabene et al. (1978) and Fiddler et al. (1978) found that Polysorbate 20 functioned adequately as an emulsifying agent in brine-cured bacon. More recently, researchers have demonstrated that α -tocopherol disperses effectively during frying of bacon slices, and therefore, application of α -tocopherol may be made by either spray or dip methods to overcome the problem of water solubility (Mergens and Newmark, 1979).

α -Tocopherol-coated salts appear to be stable since Skrypec et al. (1985) reported that their anti-nitrosamine activity was retained even after 3 months of storage at 70°F. Furthermore, α -tocopherol does not interfere with the anti-botulinal activity of nitrite (Tanaka et al., 1980). The effectiveness of α -tocopherol as an anti-nitrosamine agent,

coupled with its stability during storage, have led to the recent approval for the use of α -tocopherols in the curing ingredients (Federal Register, 1985).

Microbiology of Wiltshire Bacon

Microorganisms typical of Wiltshire bacon. Several investigators have researched the microflora associated with Wiltshire bacon (Gibbons, 1940a; Gibbons, 1940b; Gardner and Patton, 1969; Gardner, 1971). The most prevalent bacteria in mature Wiltshire bacon are micrococci (Garrard and Lochhead, 1939; Ingram, 1952; Gardner and Patton, 1969). The predominance of micrococci holds true for both the lean and fatty portions of the bacon. It has been noted that on the meaty areas of the bacon, however, gram-negative organisms such as Acinetobacter spp. and Vibrio spp. occur in greater proportions (Gardner and Patton, 1969). Other species of microorganisms which have been reported to occur in Wiltshire bacon are Clostridium, Streptococcus, Enterobacteriaceae and Alcaligenes (Ingram and Hobbs, 1954).

Microorganisms in the curing pickle. Initially, Gibbons (1940a) thought that the number of microorganisms present in the pump, cover and spent pickles affected the number of microorganisms present on the finished bacon product. However, Gibbons (1940b) later reported that the number of bacteria in the pickle is not significantly correlated with the number of bacteria on the bacon. Other factors such as contamination of the sides prior to cure (Garrard and Lochhead, 1939), age of the sides from the cure, packing practices, and shipping conditions, contribute more to the bacterial load on bacon slices than does the bacteria found in the pickle (Gibbons 1940b).

Surface sliming of Wiltshire bacon. The majority of Wiltshire bacon is cured by immersion curing. Bacon processed in this fashion is characteristically matured for 7 to 10 days after removal from the brine. This constitutes a problem since organisms which grow on the surface of the meat may predispose it to spoilage. A condition known as slime or taint of the meat may develop, slime being defined as when bacterial growth has increased to visible or tactile levels (Gibbons, 1940b). Vibriococci have been implicated as the major microorganism causing surface sliming of Wiltshire bacon (Gardner, 1975). Storage temperatures greater than 7°C and relative humidity levels greater than 90% enhance the growth of Vibriococci (Gardner, 1975). Thus, surface sliming or stickiness, discoloration and malodors can be the result of some defect in production, high air temperature and humidity, or faulty conditions of transport, storage and marketing (Gardner, 1975).

Typical bacterial count of Wiltshire bacon. Ingram (1960) reported that Wiltshire bacon sides directly out of the cure carried a surface bacterial load of 10^4 - 10^5 /cm², which increased more than ten-fold during one week of maturation at 4°C (Gardner and Patton, 1969). Similar results were obtained by Jespersen and Riemann (1958). These researchers found that microbial growth on bacon rind was greater than that found on the meat portion of the rasher, and that after 7 days of maturation at 5°C, bacterial counts of 10^5 /cm² and 10^4 /cm² were obtained for these respective areas on the bacon. A later study confirmed that the lean of the bacon does contain lower counts of bacteria than does the fat/rind portion (Gardner and Patton, 1969). Not only does the meat portion of the bacon contain the lowest numbers of bacteria, but also the rind that has been singed during the

processing of the bacon sides has a markedly reduced number of total viable bacteria. Gardner and Patton (1969) found the mean count of bacteria to be (in $\text{cm}^2 \times 10^5$) 4.5, 13.8, and 9.1 for singed rind, unsinged rind and meat portions, respectively.

Control of microbial growth on Wiltshire bacon during processing.

The constituents of the cure, and the method of curing in addition to the type of bacon have an effect on the microorganisms in Wiltshire bacon (Taylor and Shaw, 1975; Taylor et al., 1980). A hot processing method utilized on hams shows promise as an effective means of reducing bacterial counts of similarly processed meats (Barbe et al., 1966; Cornish and Mandigo, 1974).

Cure ingredients. Ingredients in the cure also affect microbial growth on Wiltshire bacon sides. Taylor and Shaw (1975) studied the effect of nitrate (5000 mg/liter) and various amounts of nitrite (250 - 2000 mg/liter) in the cure on the resultant number of bacteria on bacon. They concluded that:

- 1) Increased microbial growth during storage occurred in cures consisting of 5000 mg nitrate/liter and 1000 mg nitrite/liter, and that these effects were particularly obvious in fat and rind portions of bacon stored at 15°C.
- 2) Bacon cured in the presence of nitrate generally resulted in higher counts of lactic acid bacteria during storage than did bacon cured in the absence of added nitrate.
- 3) Bacterial counts of bacon cured with brines containing 500, 1000 or 2000 mg/liter of nitrite were initially similar. There was a general tendency for more rapid microbial growth during storage of bacon cured with brines containing 1000 or 500 mg nitrite/liter

compared to bacon cured with brine containing 2000 mg nitrite/liter.

- 4) Little difference in the total number of lactic acid bacteria was detected in bacon cured with either 1000 or 2000 mg nitrite/liter of brine during storage. However, bacon cured with only 500 mg nitrite/liter of brine showed an increase in lactic acid bacterial growth upon storage at 5 or 15°C.
- 5) Initial counts of both total viable bacteria and lactic acid bacteria were higher when the cure consisted of 250 mg nitrite/liter when compared to cure containing 2000 mg nitrite/liter, and this pattern of bacterial growth was retained during storage of bacon at 5 or 15°C.
- 6) Regardless of the cure composition, initial bacterial counts for the fat and rind portions of the bacon were similar to those counts found on the lean bacon portion. Total viable counts of the fat/rind portion remained higher than total viable counts of the lean during storage. Differences in total viable counts of the fat/rind were 10 to 100 times higher than those found on the lean portion of bacon when stored at 5 or 15°C.
- 7) Lean portions of the cured bacon invariably contained higher numbers of lactic acid bacteria than the fat/rind portion of the bacon during storage.

In summary, Taylor and Shaw (1975) found that the processing of Wiltshire bacon with various quantities of curing ingredients, or in the absence of a particular ingredient, influenced the microflora which subsequently developed during storage of the bacon. Both the relative

quantities of bacteria as well as the bacterial species can be affected.

Dry salt versus immersion curing processing. Differences in microbiological properties of Wiltshire bacon can arise from the method by which the bacon was processed. Wiltshire bacon processed with dry salt has been found to contain significantly lower ($P < 0.01$) total viable counts of bacteria than does bacon cured by the traditional Wiltshire method (Taylor et al., 1980). Table 5 illustrates typical bacteriological counts of dry salted versus immersion cured bacon sides.

Table 5. Total viable counts of Wiltshire bacon cured by two methods.^a

<u>Processing Method</u>	<u>Mean^b</u>	<u>Range</u>
Dry-salted sides	4.3	4.1 - 4.5
Immersion-cured sides	5.4	4.7 - 6.3

^aAdapted from Taylor et al. (1980)

^bTotal Viable Counts in $\log_{10}/\text{cm}^2$

Collar and back bacon. Taylor et al. (1980) reported that collar and back Wiltshire bacon showed variances in the microbial population in relation to the method of cure treatment. Vacuum packaged collar bacon prepared by dry salting tended to have lower total viable counts and lower numbers of lactic acid bacteria than bacon prepared by immersion cure. Additionally, the initial counts of total viable organisms and lactic acid bacteria for dry salted back bacon were lower than immersion cured back bacon. During storage at 5 and 15°C however,

back bacon which had been dry salt cured tended to have a greater bacterial count than similarly stored immersion cured back bacon (Tables 6 and 7). This phenomenon did not occur for collar bacon where dry cured collar bacon continued to show less microbial growth than immersion cured collar bacon. Enhanced microbial growth on dry cured back bacon may be in response to lower concentrations of curing salts than would normally be found in an immersion cured side. It was recommended that additional brine should be injected along the back of dry salt cured Wiltshire sides to compensate for the unusually low salt concentrations in back bacon (Taylor et al., 1980).

Hot processing. Rapidly processed pork has been scrutinized for its microbiological safety by several investigators (Pulliam and Kelly, 1965; Barbe et al., 1966; Mandigo and Henrickson, 1966; Barbe and Henrickson, 1967; Henrickson and Smith, 1967; Kotula, 1981). Because hot processing systems bypass the initial 24 hour chilling treatment, it was necessary to determine whether adverse microbiological conditions arose from the deletion of this processing step. Fresh uncured hams processed by the conventional or hot processing method had similar incidences of aerobes, anaerobes, and anaerobic sporeformers (Cornish and Mandigo, 1974). A comparison between cured hams, however, shows a significantly higher count ($P < 0.05$) in anaerobic bacteria processed by the conventional method in comparison to hams processed by the accelerated method (Cornish and Mandigo, 1974). Barbe et al. (1966) also observed a greater decrease in total bacterial number from fresh to cured hams processed by the rapid-processing method, as opposed to hams processed by the conventional method. According to Barbe et al. (1966), the differences of total bacterial counts for

Table 6. Total viable count of organisms on the lean of Wiltshire bacon during storage.

			Total viable count ^b				
			5°C		15°C		
Comparison	Bacon	Days Stored	Cure 1	Cure 2	Cure 1	Cure 2	
A ^c	Collar	0	4.2	5.3	4.2	5.3	
		5	---	---	5.4	6.7	
		9	4.3	6.7	6.5	6.8	
		15	5.3	6.9	7.3	7.0	
		20	6.5	6.9	---	---	
	Back	0	3.3	5.3	3.3	5.3	
		19	---	---	7.7	5.8	
		35	7.0	6.2	---	---	
	B ^d	Collar	0	5.5	6.2	5.5	6.2
			5	---	---	6.7	6.1
9			5.2	6.1	6.6	6.6	
15			5.3	5.5	7.2	6.9	
20			5.6	5.8	---	---	
Back		0	5.5	5.0	5.5	5.0	
		19	---	---	7.0	6.7	
		35	6.3	5.1	---	---	

^aAdapted from Taylor et al. (1980)

^bTotal viable count in log₁₀/g

^cDry salted with nitrate (cure 1); and immersion cured with nitrate (cure 2)

^dDry salted with nitrate (cure 1); and dry salted without nitrate (cure 2)

Table 7. Lactic acid bacteria count on the lean of Wiltshire bacon during storage.^a

			<u>Lactic acid bacteria count^b</u>				
			<u>5°C</u>		<u>15°C</u>		
Comparison	Bacon	Days Stored	Cure 1	Cure 2	Cure 1	Cure 2	
A ^c	Collar	0	2.4	1.7	2.4	1.7	
		5	---	---	4.3	5.7	
		9	2.0	5.2	5.7	5.8	
		15	4.5	5.6	6.5	6.2	
		20	5.9	6.6	---	---	
	Back	0	1.7	2.7	1.7	2.7	
		19	---	---	6.7	5.5	
		35	6.4	5.2	---	---	
	B ^d	Collar	0	2.0	1.7	2.0	1.7
			5	---	---	4.7	4.4
9			1.9	2.6	5.8	5.1	
15			4.1	3.8	6.6	6.0	
20			5.2	5.4	---	---	
Back		0	1.7	1.7	1.7	1.7	
		19	---	---	6.9	6.3	
		35	6.1	5.0	---	---	

^aAdapted from Taylor et al. (1980)

^bCount of lactic acid bacteria in log₁₀/g

^cDry salted with nitrate (cure 1); and immersion cured with nitrate (cure 2)

^dDry salted with nitrate (cure 1); and dry salted without nitrate (cure 2)

cured and uncured hams can be explained by the combination of factors associated with rapid processing; namely pre-rigor injection, higher muscle temperature, and rapidity of processing. These factors, in combination with the curing ingredients, smoking, and cooking, expose the bacteria to maximum lethal conditions, thereby significantly reducing total bacterial counts of hot processed hams (Barbe et al., 1966).

Additional factors influencing microbial growth in cured meat products. Additional factors such as nitrite level, salt concentration, pH, and storage conditions affect microbial growth on cured meat products (National Academy of Sciences, 1981). The microbiological quality of the bacon can be influenced by optimizing conditions which negatively affect microbial growth. Since the color, odor, flavor and overall appearance of the meat are the major concerns of the consumer, it is to the advantage of the processor to minimize bacterial growth on the meat product.

Nitrite as an anti-microbial agent. Nitrite has four basic functions in the curing of meat products: 1) to develop the characteristic pink color of cured meat; 2) to produce the characteristic flavor of cured meat; 3) to function as an antibotulinal agent; and 4) to act as an antioxidant (Gray, 1976).

In Europe, pork products are the most frequent cause of botulism, type B botulism being the most common offender. The frequency of Clostridium botulinum spores was found to be one spore per pound of bacon (Roberts and Smart, 1976). Although more incidences of botulism occur in Europe than in the United States and Canada, the fatality-ratio is much lower in Europe (Tompkin, 1980).

Nitrite inhibits Clostridium botulinum by preventing the outgrowth of germinated botulinal spores (Christiansen, 1980). Since nitrite concentrations in the meat decrease during storage, it is important that the level of residual nitrite be high enough to discourage spore outgrowth. It has been shown that regardless of input levels of nitrite, nitrite depletes until it levels off at approximately 5 mg/kg (Christiansen, 1980). Despite this fact, a correlation of the degree of inhibition of spore outgrowth and input levels of nitrite has been established (Christiansen et al., 1973, 1974, 1975; Hustad et al., 1973; Tompkin et al., 1977).

Nitrite is not inhibitory under anaerobic conditions and at acidic pH levels (4.5 - 5.5). A ten fold increase in the amount of nitrite present in the meat is necessary to inhibit spore outgrowth at each pH unit change over the range of 5.5 to 7.0 (Holley, 1981). A pH value of approximately 4.6 is considered to be inhibitory to C. botulinum in cured meats regardless of other cure ingredients (Christiansen, 1980). It should be noted, however, that increased nitrite depletion is observed at lower pH values (Nordin, 1969). To overcome this problem, an attempt to gradually decrease nitrite has been sought, since an initial microbial inhibition caused by the presence of nitrite, followed by a lowered pH in response to the presence of lactic acid bacteria, is an effective means of controlling spore outgrowth of Clostridium botulinum (Christiansen, 1980). The importance of pH and nitrite levels in inhibiting Clostridium botulinum spore outgrowth in summer sausages has been researched by Christiansen et al. (1975).

Although the exact mechanism(s) of the action of nitrite on the inhibition of Clostridium botulinum is not known, it has been suggested that nitrite interferes with iron or an iron-containing compound such as ferredoxin, thereby impairing energy yielding reactions (Holley, 1981). Other antibotulinal mechanisms of action of nitrite have been proposed: Nitrite (1) could produce a substance capable of inhibiting spore outgrowth when heated; (2) could act as either an oxidant or reductant in cellular biochemical reactions within spores or vegetative cells; (3) could limit substrate transport of the cell surface membrane, interfering with cellular metabolism; and (4) could interfere with cellular metabolism and repair by restricting the availability of iron (Benedict, 1980). Chelating agents such as ascorbate, isoascorbate, EDTA, sorbate and cysteine enhance the function of nitrite in inhibiting Clostridium botulinum spore outgrowth (Benedict, 1980; Christiansen, 1980; Holley, 1981). It is thought that the action of these agents in conjunction with nitrite is to chelate iron, thereby interfering with some essential iron-containing compound such as ferredoxin, which impairs energy yielding reactions (Holley, 1981).

Salt as an anti-microbial agent. Spore germination is inhibited by sodium chloride, whereas spore outgrowth is inhibited by nitrite. It has also been established that heat-damaged spores are inhibited by NaCl more readily than unheated spores (Pivnick et al., 1970). Many shelf-stable canned meat products containing minimal or no nitrite rely on the action of NaCl on heat damaged Clostridial spores (Pivnick et al., 1970). The amount of salt in the aqueous portion of the meat (brine concentration) and not the actual salt concentration in the meat determines the effectiveness of botulinal inhibition (Cerveney, 1980).

At a brine concentration of about 9.0%, toxin formation is inhibited. Meat containing lower amounts of salt may allow the formation of the toxin, yet appear organoleptically safe (Greenberg et al., 1958). Cervený (1980) has stated that under abusive conditions, cured meats produced today, with the exception of bologna and frankfurters, have more of a potential to produce toxin than if they would have been produced in 1932.

Residual nitrite concentrations are lowered in the presence of salt (Lee and Cassens, 1980). A reduction in the nitrite level may be explained by the fact that salt solubilizes the muscle proteins and aids in the extraction of these proteins from their cellular components. Nitrite probably interacts with the released proteins, seemingly lowering the level of residual nitrite.

Microbiological effects of water activity. A more important function of salt may be by decreasing the amount of available water in the meat product. Water activity (A_w) directly influences the growth of microorganisms by interfering with their metabolic activity, reproduction and resistance to environmental conditions (Leistner et al., 1981). A high A_w is therefore required for microorganisms to grow in food and a decrease in A_w would reduce the ability of the organisms to grow and multiply (Leistner et al., 1981). All phases of bacterial growth are affected by the A_w of the media, including the lag, log and death phases of microbial growth (Troller and Christian, 1978). It has been reported that a water activity above 0.935 and a salt concentration of less than 10% are the optimum requirements for the growth of Clostridium botulinum (Benedict, 1980).

The effects of pH on microorganisms. As stated earlier, the pH of food plays an important role in its antimicrobial properties. Nitrite is dissipated at lower pH levels, and it is more effective in inhibiting spore outgrowth at pH levels of 4.5 - 5.5 (Holley, 1981). The use of starter cultures containing lactic acid bacteria is becoming an increasingly popular means of lowering the pH of the meat product. The lowered pH of the meat through the use of starter cultures for bacon systems has several advantages; a desirable acid flavor from the presence of lactic acid bacteria, protection against spoilage, and decreased levels of N-nitrosamines produced at the time of frying (Bacus, 1979; Tanaka et al., 1985).

Storage conditions in relation to antimicrobial properties. Probably the single most important feature of controlling the safety of cured meats is storage temperature (Holley, 1981). Optimum temperatures for growth of C. botulinum are between 25 - 35°C, with a range of 5 - 45°C (Benedict, 1980). The antimicrobial relationships between nitrite, salt and pH are similar at 25 and 37°C. At temperatures of 20°C and less, the minimum concentrations of the above factors necessary for inhibition are much decreased (Ingram, 1973).

It has been reported that even in the absence of nitrite or sorbate, bacon inoculated with 1,300 spores of Clostridium botulinum/g and stored at 13°C for 180 days did not become toxic (Pierson, 1978). Similarly, liver sausage containing no nitrite and stored at 15°C for 14 days, did not become toxic when inoculated with 1,000 spores/g. However, the liver sausage did become toxic when stored at 20 and 25°C (Ala-Huikku et al., 1977). Additional studies by Christiansen et al.

(1973; 1974), found that ham or bacon inoculated with > 4,000 spores/g of Clostridium botulinum type A and B spores did not become toxic when stored at 7°C for 84 to 180 days. Even the control samples containing no nitrite did not become toxic over the entire storage time.

MATERIAL AND METHODS

Material

Wiltshire bacon. All pigs used in this study were obtained from the swine farm at Michigan State University (East Lansing, MI). Pigs weighed approximately 60 - 80 kg at the time of slaughter. The manufacture of Wiltshire bacon was under carefully controlled processing conditions at the Meat Laboratory at Michigan State University.

Salt. Alberger® Fine Flake salt and Alberger® Fine Flake salt coated with 3% α -tocopherol were obtained from Diamond Crystal Salt Company, St. Clair, MI.

Packaging material. Three mil pouches composed of one layer of polyvinylidene chloride (PVDC) coated on both sides with ethylene vinylacetate (EVA) were obtained from Koch, Kansas City, MO. The oxygen transport rate of these bags at 4°C was 9 ml/m²/24 hours.

Fatty acid methyl ester standards. Standards for fatty acid analyses were obtained from Larodan Fine Chemicals AB (Sweden).

Sodium nitrite. Sodium nitrite was obtained from Fisher Scientific, Detroit, MI.

Microbiological media. All media used in the analysis of microorganisms were obtained from Difco Laboratories, Detroit, MI.

All other reagents and chemicals used in the study were reagent grade.

Methods

Bacon processing procedures. Two studies were conducted to evaluate the effect of α -tocopherol-coated salt as an ingredient in the curing mixture. The initial study (Experiment A) evaluated the N-nitrosamine levels as well as the chemical, sensory and microbiological characteristics of Wiltshire bacon processed by the dry salt method and that cured by the traditional Wiltshire process. Experiment B duplicated the comparison of dry salt and immersion curing techniques except that α -tocopherol-coated salts were incorporated into the curing pickles. A total of four pigs were used for each of the 2 experiments.

For the α -tocopherol study, the α -tocopherol-coated salts were blended with regular salt so that the ingoing level of α -tocopherol in bacon was 500 mg/kg at the target salt level.

After slaughter, the sides were held overnight at 1°C. The sides were then dressed for Wiltshire curing as follows: The carcasses were split in half lengthwise, and the heads, backbones, aitchbones, tenderloins, skirts and ribs were removed. The forelegs and hindlegs were trimmed at the knees and hocks, respectively. The weight of each side was taken after trimming. At this point, the sides were labeled as to which curing treatment they were to receive. To decrease variation between pigs, the left side of each was processed by the immersion technique while the right side was dry salt-cured. Sides were designated as A, C, D and E (L if immersion cured; R if dry salted) for the control treatment (Experiment A), and Q, R, S and T (L and R as before) for the α -tocopherol-treated sides (Experiment B).

Control Wiltshire sides were injected with either brine prepared for the dry cured (right side) or immersion cured (left side) bacon to

an average weight gain of 10% (Table 8). At this point, those sides processed by the dry cure method received 3% salt (by weight), which was rubbed by hand onto the surface of the side. These sides were then hung by the achilles tendon for 5 days at 5°C.

Immersion cured sides were immersed for 3 days in freshly made brines immediately after injection (Table 8). After the immersion stage, the sides were hung by the achilles tendon for 7 days at 5°C for maturation.

Table 8. Brine formulations for Wiltshire bacon cured without α -tocopherol.

Target Level Brine	Ingredient	in Bacon	Brine Level
<u>Dry Cure Pump</u>	Na ascorbate	250 mg/kg	2,500 mg/kg
	Nitrite	156 mg/kg	1,560 mg/kg
	NaCl	1.6%	16%
	Sugar	0.1%	1.0%
<u>Immersion Pump</u>	Na ascorbate	250 mg/kg	2,500 mg/kg
	Nitrite	70 mg/kg	700 mg/kg
	NaCl	1.6%	16%
	Sugar	0.1%	1.0%
<u>Immersion Pickle</u>	Nitrite	150 mg/kg	1,500 mg/kg
	NaCl	2.3%	23%

Wiltshire sides cured with α -tocopherol-coated salts were treated in the same manner, where the left side received a brine injection

corresponding to the immersion cured brine formulation, and the right side of the pig was injected with brine formulated for dry salt curing (Table 9). Both of these brines were injected into the appropriate sides to an average weight gain of 10% as in the control experiment. Subsequent processing steps followed those outlined in the control experiment with the exception of the brine formulation differences described in Table 9.

Table 9. Brine formulations for Wiltshire bacon cured with α -tocopherol.

Target Level Brine	Ingredient	in Bacon	Brine Level
<u>Dry Cure Pump</u>	α -Tocopherol	500 mg/kg	5,000 mg/kg
	Na ascorbate	250 mg/kg	2,500 mg/kg
	Nitrite	156 mg/kg	1,560 mg/kg
	NaCl	1.6%	16%
	Sugar	0.1%	1.0%
<u>Immersion Pump</u>	α -Tocopherol	500 mg/kg	5,000 mg/kg
	Na ascorbate	250 mg/kg	2,500 mg/kg
	Nitrite	70 mg/kg	700 mg/kg
	NaCl	1.6%	16%
	Sugar	0.1%	1.0%
<u>Immersion Pickle</u>	Nitrite	150 mg/kg	1,500 mg/kg
	NaCl	2.3%	23%

Slicing, packaging and storage of bacon. Because of the differences in time of the maturation period required for dry salt and immersion cured sides, only the sides from one processing method were sliced and packaged on one day. The evening before the sides were to be sliced, they were rinsed with water to disperse any salt pockets or residual brine ingredients and returned to the cooler. The temperature of the cooler was lowered to 1°C to allow tissue hardening for ease of slicing. Collar and back portions of the Wiltshire side were sliced to approximately 3 mm thickness, randomly placed in plastic pouches according to the bacon side number and treatment, and sealed under vacuum (25 " Hg).

Bacon samples for both the collar and back portions of each side were packaged in various weight quantities which would be sufficient for the analyses to be performed. Thus, for each side, bacon was packaged as follows: 9 packages containing 25 ± 0.1 g of bacon, 3 packages containing 170 g of bacon, and 9 packages of >200 g of bacon were prepared for both the collar and back portions of the pig side. These quantities of bacon were necessary for the microbiological analyses, N-nitrosamine analyses and the sensory/TBA/nitrite/salt and other analyses, respectively. This packaging arrangement allowed for ease of sample procurement at the time of bacon analysis.

Immediately following bacon processing, a sample of collar and back bacon for each side and type of treatment (i.e., dry salt cured, immersion cured) was placed aside to be used for Day 0 analyses. The remaining pouches were divided equally and stored at either 5 or 15°C for subsequent chemical analyses.

Experimental design for Wiltshire bacon analyses. Bacon prepared for the control study was evaluated initially for N-nitrosamines and pH, and again after 20 days of storage at 5 and 15°C. Other chemical analyses of the control study occurred on Days 5, 10, 15 and 20. Bacon was analyzed at these designated times for residual nitrite concentration, fatty acid profiles, sodium chloride content, and TBA values. Microbial growth was also monitored over time at the storage conditions listed above.

Chemical analyses for the α -tocopherol-treated bacon sides were performed on Days 5, 15 and 20. These analyses were identical to those performed for the control bacon samples, in addition to a random determination of the residual α -tocopherol content. N-Nitrosamine content and pH values were obtained on Day 0 and Day 21.

Sensory. The bacon was also evaluated by a taste panel using a hedonic rating procedure (Appendix 1). Results from the taste panel scores were compared with TBA values taken from identically stored bacon. An ANOVA statistical computation was performed on TBA values from bacon samples stored for 15 days at 5°C. Flavor acceptability and overall evaluation were also statistically analyzed by ANOVA for collar and back bacon stored for the same period of time at 5°C.

The taste panel consisted of at least 10 untrained members, who rated the samples in degrees of perception of the following bacon qualities: flavor acceptability; color intensity; texture; saltiness; and overall acceptability. The panelists were served 2 to 8 samples per sitting. At the time of the testing session, the panelists were served a plate containing no more than 4 samples contained in separate plastic sampling cups. Each sample was identifiable through a randomly

selected 3-digit number. At no time did the panelists know which samples were being served. The samples consisted of collar and back bacon, dry salt and/or immersion cured, which had been stored as stated above.

The bacon was cooked in a manner similar to that described by Taylor et al. (1975). Immediately before serving, the bacon was reheated in a microwave oven. Panelists were given unsalted crackers and water to clear their palates between samples.

N-Nitrosamine analysis. To determine the differences in volatile N-nitrosamine contents of bacon cured by dry salt and immersion techniques, N-nitrosamine analyses of the bacon samples were performed initially and at the end of the storage periods.

Microbiological examination of bacon sides. To monitor the growth of microbes, total viable counts (TVC) of the bacon sides were performed during various stages of processing. For dry salt cured sides, the TVC was taken after pumping the sides with brine, and also after the 5 day maturation period. Immersion cured sides were tested for TVC following brine injection, following the 3 day immersion step, and finally after the 7 day maturation period.

Microbiological evaluation over time. The control bacon, i.e. without α -tocopherol, was assayed for several microbiological parameters after storage at 5 and 15°C. The conditions of the microbiological evaluation are listed in Table 10.

Table 10. Conditions for microbiological assay of Wiltshire bacon.

Isolation of Microbes	Media Used	Incubation Temperature (°C)	Hours of Incubation
Total Plate Count	Plate Count Agar	32	48
Psychrotroph Count	Plate Count Agar	7	120
Lactobacillus Agar Count	Lactobacillus Agar	32	48
Halophile Count	Plate Count Agar + 4% NaCl	32	48

Methods of Analysis

Bacon frying. Two teflon coated electric skillets (Sears Roebuck, Inc.) were used for the frying of bacon. Frying was modeled after the procedure outlined by Mottram et al. (1977). Briefly, the skillets were preheated to $178 \pm 3^{\circ}\text{C}$ (352°F) and allowed to cycle at least 10 minutes. Strips of bacon were placed onto the hot skillet so that the strips were not overlapping and enough room remained for turning. Bacon strips were fried for a total of 12 minutes (6 minutes/side). After frying, the bacon was removed to paper towels where the excess fat was allowed to drain off. The fried bacon was wrapped in aluminum foil and stored overnight in a freezer set at -20°C .

The cooked-out fat remaining in the skillet was poured into containers and reserved for N-nitrosamine analysis. The skillet was wiped free of residual fat with paper towels between fryings of the same type of bacon. If bacon of a different type or treatment was fried, the skillet was washed out before frying.

Quantitation of N-nitrosamines in bacon and cooked-out fat. The fried bacon was frozen, ground and analyzed for N-nitrosamines by a gas chromatography-thermal energy analyzer (GC-TEA) procedure described by Reddy et al. (1982). This procedure included the addition of 1 g of ammonium sulfamate to the distillation flask immediately before the onset of distillation to decrease the possibility of artifactual formation of N-nitrosamines during sample preparation.

Quantitation of the volatile N-nitrosamines recovered from the bacon samples was carried out using a GC-TEA system comprised of a Varian 3700 gas chromatograph coupled to a TEA Model 502 LC (Thermal Electron Corp., Waltham, MA) via a 1/8" glass-lined stainless steel transfer line. The GC column was a stainless steel column (6' x 1/8" i.d.) packed with 10% Carbowax 20 M + 5% KOH on 80/100 mesh Chromosorb W (Supelco Inc., Bellefonte, PA). The operating conditions were as follows: nitrogen flow rate was 30 ml/min; initial temperature was 140°C which was held for 1 minute; the rate of temperature change was 1°C/min for 15 minutes, resulting in a final temperature of 180°C which was held for 2 min. The injector temperature was 150°C, and the TEA pyrolyzer furnace was set at 425°C. The reaction chamber pressure of the TEA was 1.5 Torr, and the GC-TEA transfer line was heated to 200°C.

N-Nitrosamines in the cooked-out fats were determined by the method described by Owens and Kinast (1980) with the exception that 1 g of ammonium sulfamate was added to the distillation flask before the onset of distillation. Quantitation was as described for the bacon samples.

Chemical analyses. Moisture, fat, salt, pH and residual nitrite determinations were performed according to standard AOAC procedures (1984).

Thiobarbituric acid test (TBA). Uncooked ground collar and back bacon samples were analyzed by the 2-thiobarbituric acid (TBA) procedure of Tarladgis et al. (1960), as modified by Zipser and Watts (1962) for meat samples containing nitrite. TBA values were expressed as mg malonaldehyde/kg of bacon. Statistical analysis of the data was accomplished by the procedure of ANOVA analysis (Gill, 1978).

Fatty acid analysis. The fatty acid composition of the bacon adipose tissue was determined by the methylation procedure of Morrison and Smith (1964). Analysis of the fatty acid methyl esters was performed on a Hewlett Packard gas chromatograph (Model 5840A) equipped with a flame ionization detector (FID) and a Hewlett Packard 18850A GC integrator. The glass column (2 m x 2 mm. i.d.) was packed with 15% diethylene glycol succinate (DEGS) on Chromasorb W 60/80 mesh. Operating conditions are as follows: The GC carrier gas, nitrogen, was set at a flow rate of 30 ml/min; the injection port temperature was 210°C; the column temperature was isothermal and set at 190°C; the FID temperature was 350°C. The identification of the fatty acid methyl ester peaks was determined by comparison of retention times of fatty acid methyl ester standards which were assayed under identical conditions.

α -Tocopherol analysis by high performance liquid chromatography. Bacon samples from Experiment B were analyzed for their α -tocopherol content according to the procedure of Thompson and Hatina (1979). The α -tocopherol analyses were carried out using a Waters liquid chromatograph (ALC-201 Model) equipped with a U6K loop injector, Model 440 UV absorption detector and a micro-Porasil column P/N 27477 S/N (Waters Associates, Inc., Milford, MA). The UV detector was set at 280 nm.

The solvent system consisted of 85% hexane/15% chloroform at a flow rate of 2 ml/min.

Total viable count of microorganisms. Total viable counts were taken in a manner similar to that described by Taylor et al. (1980). Five sterile cotton swabs were swabbed over five/10 cm² areas of the rind or pleura portion of the bacon side, then bulked into 10 ml of 0.1% peptone. The swabs thus represented a 50 cm² composite sample of the rind or pleura of the bacon side. The bulked swabs were held at 4°C until plated, which occurred within 2 hours of sampling. Samples were plated in duplicate onto 4% PCA + NaCl agar at dilutions of 10⁻¹, 10⁻² and 10⁻³. Plated samples were incubated at 32°C and the total viable count obtained after 2 days. All microbiological evaluations utilized the pour plate method.

Microbial enumeration of stored bacon. At the time of microbial evaluation, the preweighed bacon samples in vacuum pouches were examined for leaks. The packs were opened aseptically and the contents homogenized in 225 ml of 0.1% peptone using a stomacher blender. The homogenate was then plated in duplicate using appropriate agars and serial dilutions. After plating, the plates were incubated as described in Table 10. Microbial enumeration of the stored bacon was performed as described by the AOAC (1970) and APHA (1972).

RESULTS AND DISCUSSION

Processing weights, moisture, fat and salt contents of Wiltshire bacon. During the processing of Wiltshire bacon, it was necessary to measure pig side weight in order to inject the sides to approximately 110% of their green weight with brine. The target weight gains, actual weight gains, and also the weights of the trimmed bacon sides are listed in Table 11. Results of proximate analyses and pH values of the bacon at the onset and completion of the experiments are summarized in Tables 12, 13, and 14. The moisture contents in the cured sides of both the control and α -tocopherol studies are somewhat lower than those cited by Jolley (1979) and Taylor et al. (1982). This difference is probably attributable to the sampling methods, since these researchers performed their moisture analysis on lean trimmed of extraneous fat. The pH values of the bacon samples agree with those previously reported for cured Wiltshire bacon sides (Taylor and Shaw, 1975; Jolley, 1979; Taylor et al., 1980; Taylor et al., 1982).

The salt levels in Wiltshire bacon sides prepared with and without α -tocopherol-coated curing salts are summarized in Tables 15 and 16. It appears that salt uptake by the bacon is not favored by the type of bacon being cured (back vs collar bacon). Taylor et al. (1980) found that dry cured back bacon did not have as high a salt concentration as did dry cured collar bacon or immersion cured back bacon. They suggested that the salt concentration of dry cured back bacon could be increased by injecting more cure into that area during processing.

Table 11. Pig side weights after trimming and actual weight gains of bacon sides after brine injection.

Side ^{a,b} Identification	Trimmed Weight (kg)	Target Weight (kg)	Actual Weight (kg)
A-R	23.0	25.3	25.5
C-R	24.6	27.0	26.9
D-R	26.0	28.6	28.7
E-R	23.0	25.3	25.3
A-L	23.4	25.8	25.7
C-L	25.9	28.4	28.4
D-L	25.5	28.1	28.3
E-L	23.2	25.5	25.5
Q-R	21.7	23.9	24.2
R-R	21.0	23.1	23.0
S-R	22.5	24.7	24.7
T-R	22.3	24.5	24.7
Q-L	21.0	23.0	23.5
R-L	21.4	23.5	23.5
S-L	23.4	25.8	26.0
T-L	21.8	23.9	24.0

^aR = right side; L = left side

^bPigs A, C, D, E used for control study (no α -tocopherol)

Pigs Q, R, S, T used for α -tocopherol study

Table 12. Moisture and fat contents of control Wiltshire bacon.

Treatment	Days of Storage	Temperature of Storage (°C)	Moisture ^a (%)	Fat ^a (%)
<u>Immersion Cured</u>				
back	0	-	47.6 $\pm$ 3.9	35.2 $\pm$ 6.8
collar	0	-	51.9 $\pm$ 3.6	32.8 $\pm$ 1.8
back	20	5	42.0 $\pm$ 1.5	35.0 $\pm$ 0.2
collar	20	5	50.9 $\pm$ 7.2	33.6 $\pm$ 6.7
back	20	15	47.3 $\pm$ 2.1	32.1 $\pm$ 1.9
collar	20	15	53.1 $\pm$ 1.9	28.0 $\pm$ 0.6
<u>Dry Salt Cured</u>				
back	0	-	44.7 $\pm$ 3.5	37.9 $\pm$ 6.5
collar	0	-	52.3 $\pm$ 4.9	24.3 $\pm$ 3.0
back	20	5	46.9 $\pm$ 3.6	37.4 $\pm$ 2.3
collar	20	5	51.0 $\pm$ 7.0	26.0 $\pm$ 3.2
back	20	15	45.6 $\pm$ 3.0	32.5 $\pm$ 1.4
collar	20	15	57.6 $\pm$ 4.2	19.3 $\pm$ 1.1

^a Moisture and fat determinations were performed in duplicate.

Table 13. Moisture and fat contents of Wiltshire bacon cured with α -tocopherol.

Treatment	Days of Storage	Temperature of Storage ($^{\circ}$ C)	Moisture ^a (%)	Fat ^a (%)
<u>Immersion Cured</u>				
back	0	-	56.5 $\pm$ 1.1	23.4 $\pm$ 0.6
collar	0	-	60.5 $\pm$ 5.8	24.2 $\pm$ 4.9
back	20	5	62.0 $\pm$ 1.6	15.2 $\pm$ 1.6
collar	20	5	60.4 $\pm$ 2.0	18.8 $\pm$ 2.2
back	20	15	59.4 $\pm$ 2.7	17.7 $\pm$ 2.7
collar	20	15	57.5 $\pm$ 1.5	25.0 $\pm$ 0.1
<u>Dry Salt Cured</u>				
back	0	-	52.2 $\pm$ 2.2	27.4 $\pm$ 0.9
collar	0	-	56.0 $\pm$ 3.3	24.4 $\pm$ 5.8
back	20	5	57.3 $\pm$ 2.7	20.1 $\pm$ 1.0
collar	20	5	57.5 $\pm$ 5.8	26.8 $\pm$ 3.5
back	20	15	NA ^b	NA
collar	20	15	NA	NA

^a Moisture and fat determinations were performed in duplicate.

^b NA=Data not available due to limitations of sample numbers.

Table 14. pH values for Wiltshire bacon processed with and without α -tocopherol.

Sample Treatment		Control Bacon	α -Tocopherol Bacon
<u>Immersion Cured</u>			
Day 0 back		5.84	5.72
Day 0 collar		6.02	5.80
Day 20 back	5°C	5.88	5.77
Day 20 collar	5°C	6.10	5.79
Day 20 back	15°C	5.85	5.77
Day 20 collar	15°C	6.05	5.75
<u>Dry Salt Cured</u>			
Day 0 back		5.69	5.73
Day 0 collar		5.85	5.65
Day 20 back	5°C	5.82	5.70
Day 20 collar	5°C	6.04	5.72
Day 20 back	15°C	6.02	5.85
Day 20 collar	15°C	5.91	5.80

Table 15. Salt content of control Wiltshire bacon samples stored at 5 and 15°C.^a

Percent salt						
<u>Dry Salt Cured</u>						
<u>Day</u>	<u>5°C</u>	<u>backs</u>	<u>15°C</u>	<u>5°C</u>	<u>collars</u>	<u>15°C</u>
0		3.23			3.67	
5	2.95		3.20	4.08		3.13
10	3.42		3.48	3.76		3.16
15	3.72		3.52	3.68		3.82
20	3.54		2.98	3.99		3.40
<u>Immersion Cured</u>						
<u>Day</u>	<u>5°C</u>	<u>backs</u>	<u>15°C</u>	<u>5°C</u>	<u>collars</u>	<u>15°C</u>
0		3.17			3.00	
5	3.93		3.48	3.66		3.42
10	3.63		3.44	3.29		3.65
15	3.22		3.26	3.28		3.18
20	3.83		4.11	3.10		3.45

^aSalt levels were determined in duplicate for each bacon sample.

Table 16. Salt content of α -tocopherol-treated Wiltshire bacon samples stored at 5 and 15°C.^a

Percent salt					
<u>Dry Salt Cured</u>					
<u>Day</u>	<u>backs</u>		<u>collars</u>		
	<u>5°C</u>	<u>15°C</u>	<u>5°C</u>	<u>15°C</u>	
0		3.44		4.74	
5	3.58	3.86	4.03		3.89
15	3.92	3.83	4.02		4.38
20	4.01	NA ^b	3.99		4.22
<u>Immersion Cured</u>					
<u>Day</u>	<u>backs</u>		<u>collars</u>		
	<u>5°C</u>	<u>15°C</u>	<u>5°C</u>	<u>15°C</u>	
0		4.11		3.91	
5	4.23	4.03	3.88		3.57
15	3.65	4.46	3.53		4.01
20	4.68	4.84	3.75		3.30

^aSalt levels were determined in duplicate for each bacon sample.

^bNA=Data not available due to limitations of sample numbers.

Results from this study indicate that increased salt concentrations of dry cured Wiltshire back bacon is possible through the additional application of salt in the back bacon area of the side during processing.

Residual nitrite levels in Wiltshire bacon. Residual nitrite concentrations in the uncooked bacon samples during storage (control study) were determined and are presented in Figures 3 and 4. Individual data are presented in Appendix 2. On Day 0 (i.e., immediately after packaging), back bacon samples prepared by immersion curing contained an average nitrite concentration of 105.9 mg/kg. Dry salted back samples had an average nitrite level of 55.7 mg/kg. Collar bacon samples at Day 0 contained average nitrite levels of 93.9 and 67.1 mg/kg for the immersion cured and dry cured bacon, respectively. Taylor et al. (1980) reported very similar initial nitrite levels for dry salted back and collar bacon and immersion cured back bacon, although they reported a higher initial nitrite concentration for immersion cured collar bacon.

Residual nitrite in the backs and collars in general, decreased gradually over time until a value of less than 50% of the initial nitrite concentration was reached at Day 20. Similar nitrite depletion patterns were observed by Taylor and Shaw (1975) and Taylor et al. (1980, 1982). In the present study, the nitrite levels in the collar bacon when stored at 5°C for 20 days were somewhat higher than expected. However, this trend could be due to the random sampling procedures applied in the study.

Additionally, as shown in Figures 3 and 4, there was a more rapid depletion of nitrite resulting in lower final concentrations when the

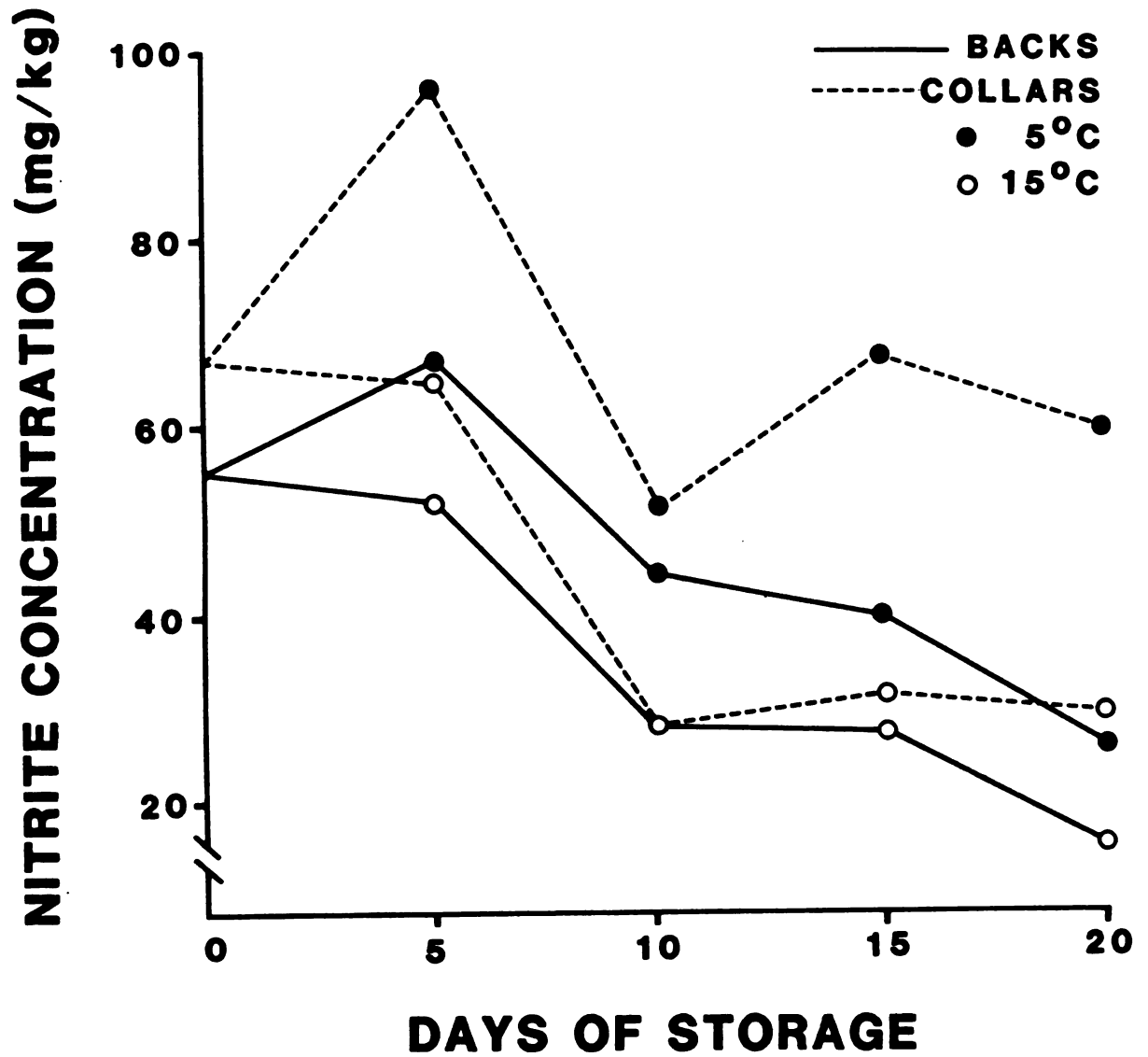


Figure 3. Nitrite depletion in dry salt-cured Wiltshire bacon during storage at 5°C and 15°C.

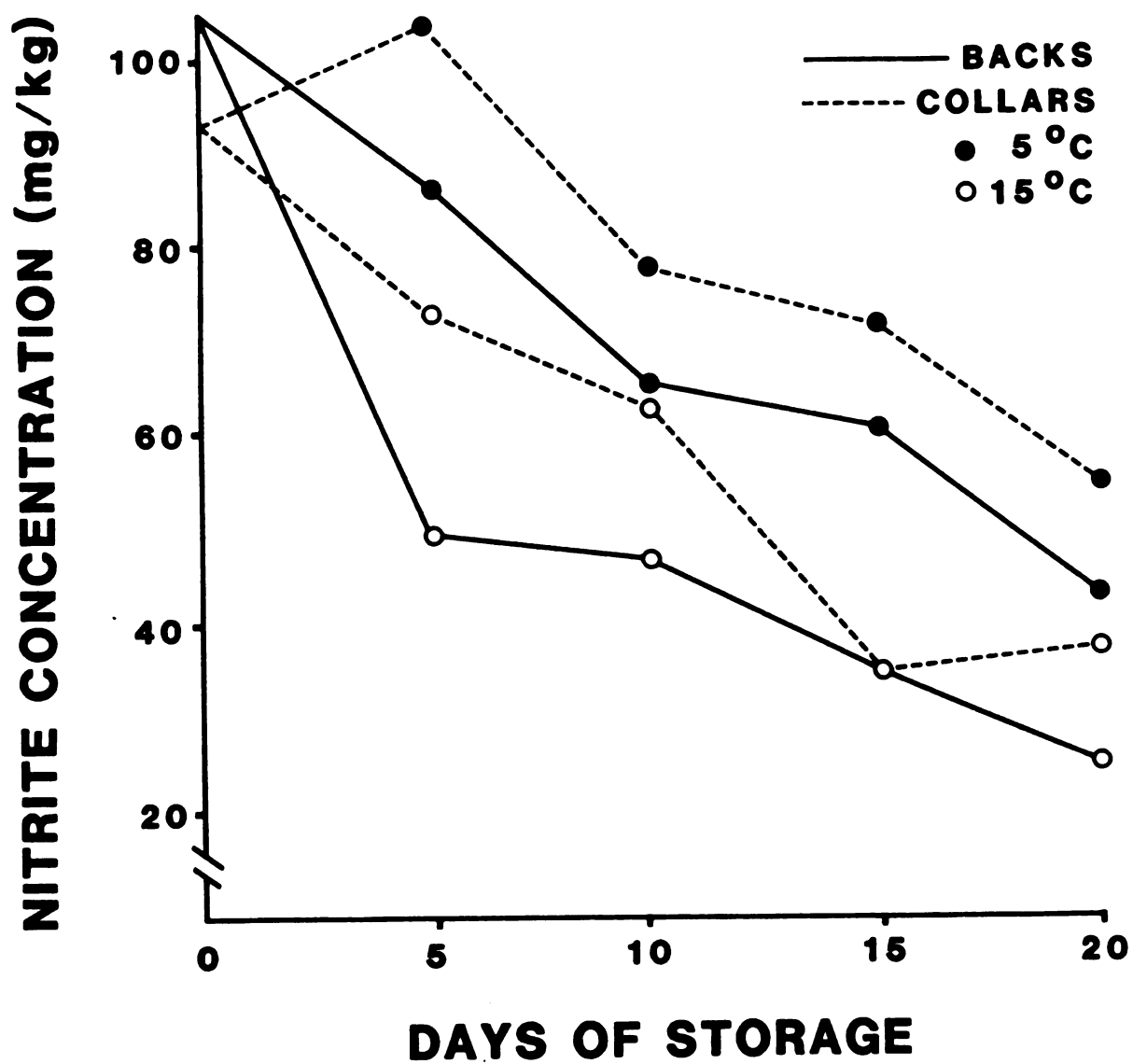


Figure 4. Nitrite depletion in immersion-cured Wiltshire bacon during storage at 5°C and 15°C.

bacon samples were stored at the higher temperature (15°C). Taylor and Shaw (1975) and Taylor et al. (1980) also observed a greater rate of nitrite depletion when bacon was stored at elevated temperatures (15°C). It has been noted that less than 50% of the added nitrite in meat systems can be accounted for by chemical analysis after the completion of processing (Cassens et al., 1974). The fate of nitrite at present is not completely resolved, but the reactions of nitrite with the meat components account for the low percent recovery of the ingoing nitrite levels (Cassens et al., 1979). Elevated temperatures are also known to be responsible for the loss of nitrite in cured meat products during processing and storage (Cassens et al., 1979). Nevertheless, a typical rate of nitrite depletion for meat systems having ingoing nitrite levels of 156 and 50 mg/kg results in a level of nitrite of approximately 5 mg/kg after 21 days of storage at 0°C (Christiansen, 1980). Although only a very small fraction of the added nitrite is detectable as volatile N-nitrosamines in cured meat, the quantity of residual nitrite remaining in the meat plays a key role in the amount of N-nitrosamines formed (Cassens et al., 1979; Dudley, 1979; Sebranek, 1979).

Nitrite depletion patterns for Wiltshire bacon cured with α -tocopherol-coated salts are represented in Figures 5 and 6, and in Appendix 3, and show similar trends as for the control bacon samples. An increase in the residual nitrite concentration was reported in immersion-cured α -tocopherol-treated collar bacon on Day 20 of bacon stored at 5°C. This increase in residual nitrite concentration may again be due to sampling error.

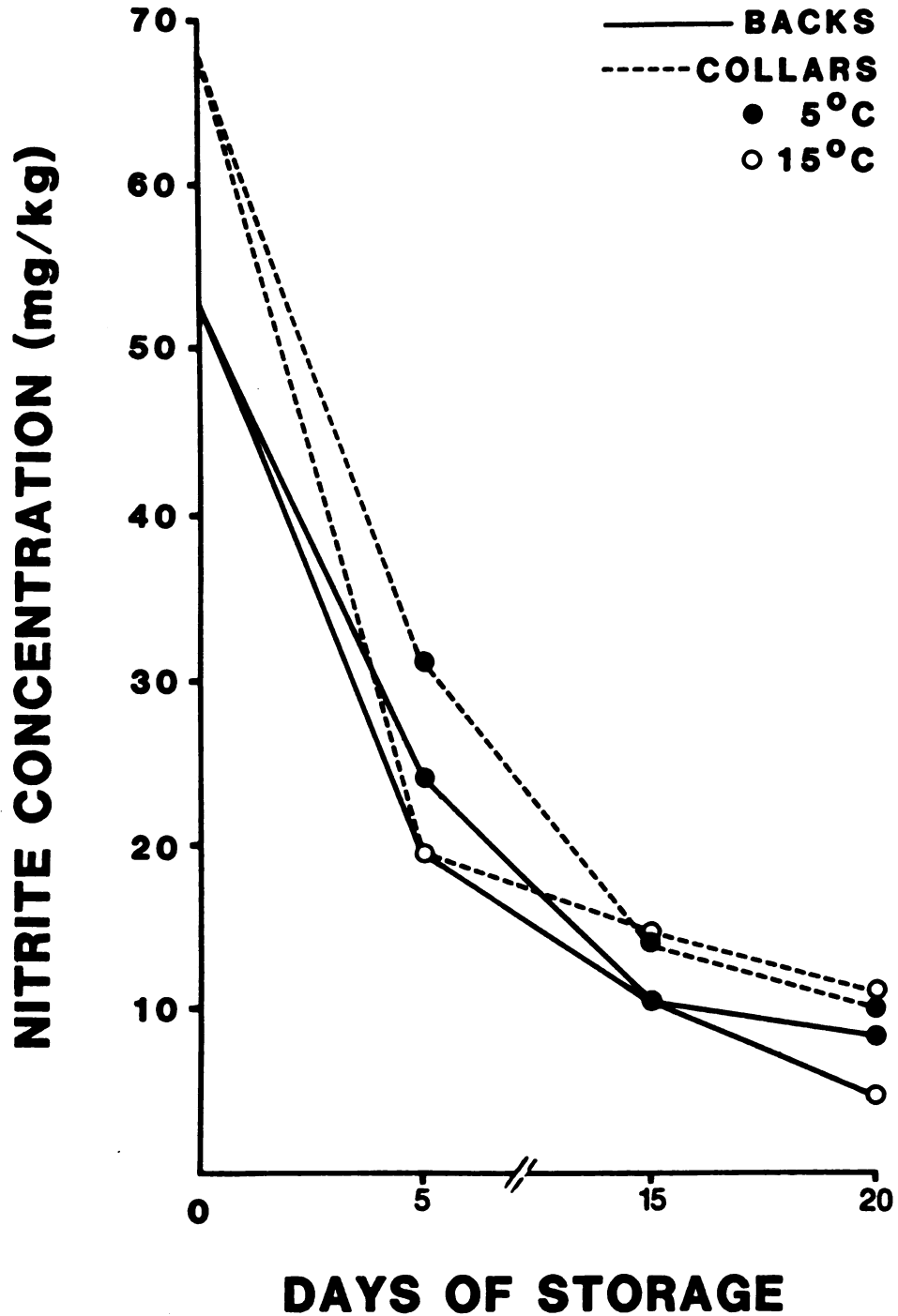


Figure 5. Nitrite depletion in dry salt-cured Wiltshire bacon containing α -tocopherol during storage at 5°C and 15°C.

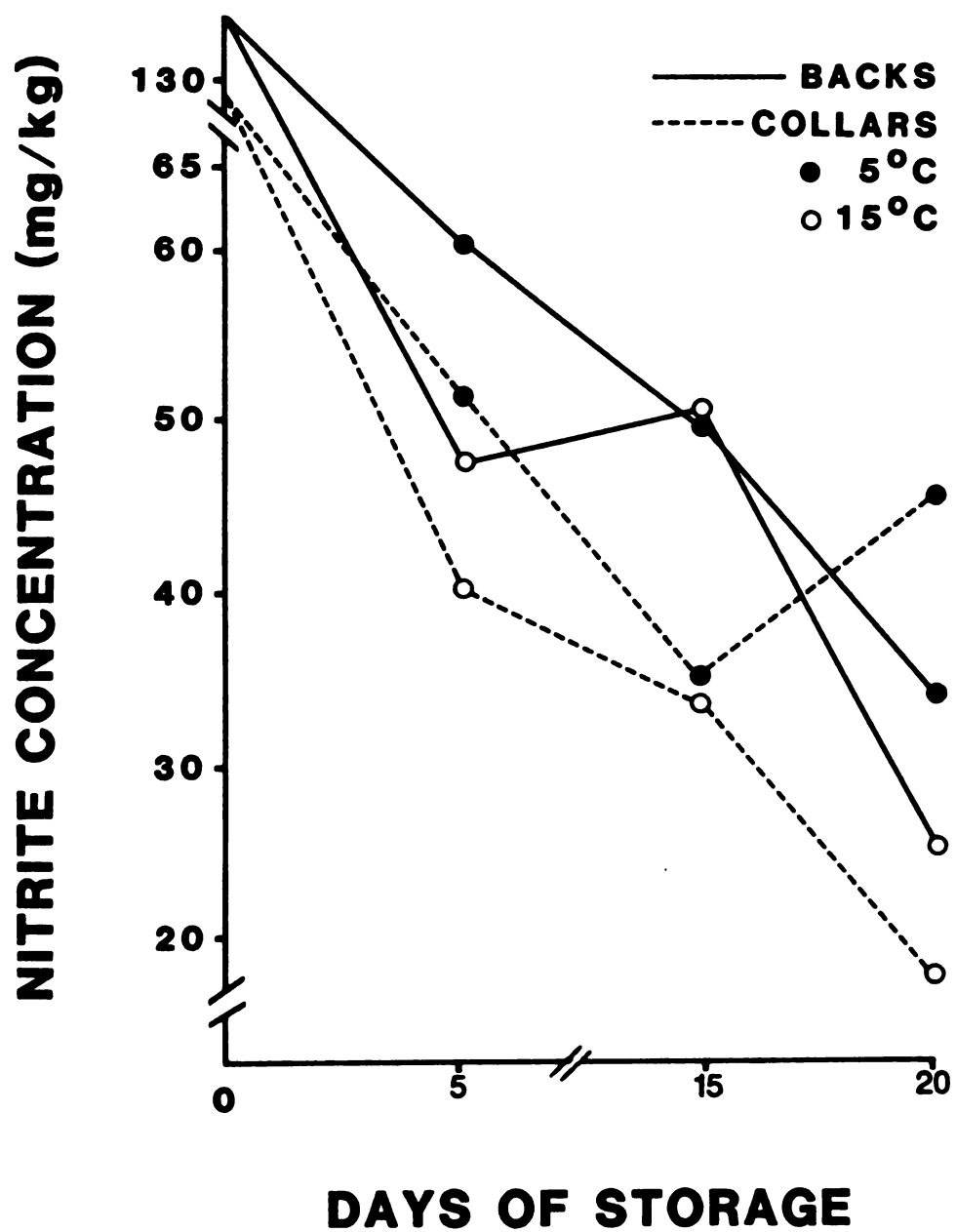


Figure 6. Nitrite depletion in immersion-cured Wiltshire bacon containing α -tocopherol during storage at 5°C and 15°C.

The residual nitrite data from this study indicate that dry salt curing of Wiltshire sides produces bacon having lower nitrite levels than bacon cured by the immersion technique. These results agree with those cited by Taylor et al. (1980). From a consumer standpoint, this is a desirable factor as long as the bacon is organoleptically and microbiologically acceptable, since high levels of residual nitrite may produce health risks (Taylor and Shaw, 1975). Wiltshire bacon which has been dry cured with nitrite and without nitrate has also been shown to be acceptable (Taylor et al., 1980; Taylor et al., 1982). The advantage of low values of nitrite in dry salted Wiltshire bacon extends to the processor, as dry cured bacon is processed more rapidly than immersion cured bacon.

N-Nitrosamines in Wiltshire bacon. Immersion-cured and dry-cured Wiltshire bacon samples were analyzed for volatile N-nitrosamines immediately after processing and packaging (Day 0). N-Nitrosamine analyses were also performed after storing the bacon samples for 20 days at 5 and 15°C. Duplicate analyses of representative samples of each bacon side were performed on both collar and back bacon. The average values and standard deviations for the control bacon samples (i.e., without α -tocopherol) are listed in Table 17. N-Nitrosamine concentrations in bacon processed with α -tocopherol-coated salts are presented in Table 18.

Comparison of N-nitrosamine levels in immersion-cured and dry-cured Wiltshire bacon. It has been reported that fried, dry-cured bacon made from pork bellies contain higher levels of N-nitrosamines than bacon produced by pumping brines (Pensabene et al., 1979b; Nitrite Safety Council, 1980). Data on the N-nitrosamine characteristics of

Table 17. N-Nitrosamine levels (ug/kg) in control Wiltshire bacon samples.^a

Bacon Sample	Day 0	Day 20 5°C	Day 20 15°C
<u>IMMERSION</u>			
<u>Back</u>			
NDMA	10.6±2.5	5.3±0.3	6.2±1.3
NPYR	22.5±12.0	7.2±1.9	12.7±2.6
<u>Collar</u>			
NDMA	11.5±1.7	5.6±1.1	4.7±2.0
NPYR	36.5±12.4	11.3±4.5	14.3±4.2
<u>DRY SALT</u>			
<u>Back</u>			
NDMA	5.0±2.6	4.9±1.6	4.9±0.7
NPYR	9.7±3.1	7.4±2.0	5.0±0.8
<u>Collar</u>			
NDMA	5.6±3.2	7.5±0.8	10.7±3.0
NPYR	7.5±1.8	8.5±4.0	9.3±3.0

^aN-Nitrosamine values represent an average of duplicate samples per bacon side, two determinations per sample.

Table 18. N-Nitrosamine levels (ug/kg) in Wiltshire bacon samples processed with α -tocopherol.^a

Bacon Sample	Day 0	Day 20 5°C	Day 20 15°C
<u>IMMERSION</u>			
<u>Back</u>			
NDMA	9.4±2.8	3.7±1.6	4.4±1.7
NPYR	5.9±4.7	4.4±4.8	7.0±5.0
<u>Collar</u>			
NDMA	10.0±5.3	3.1±1.3	2.8±1.7
NPYR	5.2±5.1	4.3±5.1	5.5±2.5
<u>DRY SALT</u>			
<u>Back</u>			
NDMA	6.4±0.9	3.1±1.4	1.6±0.3
NPYR	4.0±0.8	2.3±1.3	2.3±0.6
<u>Collar</u>			
NDMA	7.1±2.5	3.2±1.1	2.6±0.6
NPYR	4.5±1.3	2.4±1.8	1.9±0.6

^aN-Nitrosamine values represent an average of duplicate samples per bacon side, two determinations per sample.

Wiltshire bacon, however, are limited and focus primarily on the NPYR content.

Mottram et al. (1977) reported average NPYR levels of 15.6 ug/kg in fried whole rashers of immersion-cured Wiltshire bacon. The same rashers contained an average NDMA concentration of 6.7 ug/kg upon frying. Results from the present study indicate that dry-salt cured bacon contains lower NPYR and NDMA levels than bacon processed by immersion curing. NPYR levels in the control bacon samples were higher in every instance in the immersion-cured bacon compared to the dry salt-cured bacon samples, with the exception of the back bacon samples stored at 5°C for 20 days. Similarly, dry salt-cured bacon prepared with α -tocopherol coated salts contained less NPYR than their immersion cured counterparts. The levels of NDMA in both control and α -tocopherol-treated Wiltshire bacon samples are generally lower for dry-cured bacon than immersion-cured bacon. A few exceptions to this generalization exist, but this could be the result of sampling error. While dry-salt cured Wiltshire bacon appears to contain lower levels of N-nitrosamines, more samples would have to be prepared and analyzed to establish whether the decrease is significantly different.

α -Tocopherol has been shown to be an effective N-nitrosamine blocking agent by several researchers (Fiddler et al., 1978; Mergens and Newmark, 1979; Gray et al., 1982; Reddy et al., 1982). A comparison of the N-nitrosamine levels in α -tocopherol-treated bacon and control bacon indicates that α -tocopherol can effectively reduce N-nitrosamine formation in fried bacon (Tables 17 and 18). This observation was particularly noticeable in NPYR levels where up to 85.8% inhibition was realized (Table 19). N-nitrosodimethylamine

formation was also inhibited by the inclusion of α -tocopherol in the cure and inhibition values as high as 75.7% were observed.

N-Nitrosamine data and the percent inhibition by α -tocopherol are summarized in Tables 20 and 21. Bacon cured with α -tocopherol-coated salts as a curing ingredient showed an average NPYR inhibition of 64% and 63% when fried on Day 0 and after 20 days of storage at 5°C, respectively. Lower levels of inhibition were realized for NDMA where an average of 12% and 42% inhibition was encountered at Day 0 and Day 20 of a 5°C storage period. Similar inhibition values were obtained with bacon stored at 15°C over a 20 day period, where average NPYR inhibition levels of 65% and 60% were observed on Day 0 and after 20

Table 19. Percent inhibition of N-nitrosamine formation in Wiltshire bacon processed with α -tocopherol-coated salts.

Samples	Immersion Cure		Dry Salt Cure	
	<u>NDMA</u>	<u>NPYR</u>	<u>NDMA</u>	<u>NPYR</u>
<u>Backs</u>				
Day 0	11.3%	73.8%	NI ^a	58.8%
Day 20 5°C	30.2%	38.9%	36.7%	68.9%
Day 20 15°C	29.0%	44.9%	64.4%	54.0%
<u>Collars</u>				
Day 0	13.0%	85.8%	NI	40.0%
Day 20 5°C	44.6%	62.0%	57.3%	81.7%
Day 20 15°C	40.4%	61.5%	75.7%	79.6%

^aNI = no inhibition of N-nitrosamines was observed.

Table 20. Effect of α -tocopherol-coated salts on N-nitrosamine inhibition in Wiltshire bacon stored at 5°C.

Bacon Sample	NDMA (ug/kg)		NPYR (ug/kg)	
	Day 0 ^a	Day 20 ^b	Day 0	Day 20
<u>Immersion Cure</u>				
<u>Back</u>				
Control	10.6 $\pm$ 2.5	5.3 $\pm$ 0.3	22.5 $\pm$ 12.0	7.2 $\pm$ 1.9
α -Tocopherol	9.4 $\pm$ 2.8 (11) ^c	3.7 $\pm$ 1.6 (30)	5.9 $\pm$ 4.7 (74)	4.4 $\pm$ 4.8 (39)
<u>Collar</u>				
Control	11.5 $\pm$ 1.7	5.6 $\pm$ 1.1	36.5 $\pm$ 12.4	11.3 $\pm$ 4.5
α -Tocopherol	10.0 $\pm$ 5.3 (13)	3.1 $\pm$ 1.3 (45)	5.2 $\pm$ 5.1 (86)	4.3 $\pm$ 5.1 (62)
<u>Dry Salt Cure</u>				
<u>Back</u>				
Control	5.0 $\pm$ 2.6	4.9 $\pm$ 1.6	9.7 $\pm$ 3.1	7.4 $\pm$ 2.0
α -Tocopherol	6.4 $\pm$ 0.9 (--)	3.1 $\pm$ 1.4 (37)	4.0 $\pm$ 0.8 (59)	2.3 $\pm$ 1.3 (69)
<u>Collar</u>				
Control	5.6 $\pm$ 3.2	7.5 $\pm$ 0.8	7.5 $\pm$ 1.8	8.5 $\pm$ 4.0
α -Tocopherol	7.1 $\pm$ 2.5 (--)	3.2 $\pm$ 1.1 (57)	4.5 $\pm$ 1.3 (40)	2.4 $\pm$ 1.8 (82)

^aBacon was fried and analyzed immediately after packaging.

^bBacon was fried and analyzed after 20 days storage at 5°C.

^cFigures in parentheses represent percent N-nitrosamine inhibition by α -tocopherol (ingoing level, 500 mg/kg) in the bacon.

Table 21. Effect of α -tocopherol-coated salts on N-nitrosamine inhibition in Wiltshire bacon stored at 15°C.

Bacon Samples	NDMA (ug/kg)		NPYR (ug/kg)	
	Day 0 ^a	Day 20 ^b	Day 0	Day 20
<u>Immersion Cure</u>				
<u>Back</u>				
Control	10.6 $\pm$ 2.5	6.2 $\pm$ 1.3	22.5 $\pm$ 12.0	12.7 $\pm$ 2.6
α -Tocopherol	9.4 $\pm$ 2.8 (11) ^c	4.4 $\pm$ 1.7 (29)	5.9 $\pm$ 4.7 (74)	7.0 $\pm$ 5.0 (45)
<u>Collar</u>				
Control	11.5 $\pm$ 1.7	4.7 $\pm$ 2.0	36.5 $\pm$ 12.4	14.3 $\pm$ 4.2
α -Tocopherol	10.0 $\pm$ 5.3 (13)	2.8 $\pm$ 1.7 (40)	5.2 $\pm$ 5.1 (86)	5.5 $\pm$ 2.5 (62)
<u>Dry Salt Cure</u>				
<u>Back</u>				
Control	5.0 $\pm$ 2.6	4.9 $\pm$ 0.7	9.7 $\pm$ 3.1	5.0 $\pm$ 0.8
α -Tocopherol	6.4 $\pm$ 0.9 (--)	1.6 $\pm$ 0.3 (64)	4.0 $\pm$ 0.8 (59)	2.3 $\pm$ 0.6 (54)
<u>Collar</u>				
Control	5.6 $\pm$ 3.2	10.7 $\pm$ 3.0	7.5 $\pm$ 1.8	9.3 $\pm$ 3.0
α -Tocopherol	7.1 $\pm$ 2.5 (--)	2.6 $\pm$ 0.6 (76)	4.5 $\pm$ 1.3 (40)	1.9 $\pm$ 0.6 (80)

^aBacon was fried and analyzed immediately after packaging.

^bBacon was fried and analyzed after 20 days storage at 15°C.

^cFigures in parentheses represent percent N-nitrosamine inhibition by α -tocopherol (ingoing level, 500 mg/kg) in the bacon.

days of 15°C storage. Average inhibitions of 12% and 52% for NDMA were obtained on Day 0 and Day 20 of 15°C storage, respectively.

An ANOVA was performed to determine significant differences between NPYR and NDMA levels of collar or back bacon samples cured with or without α -tocopherol-coated salts. This analysis was performed on Day 20 bacon samples where a statistical difference ($P < 0.05$) was observed with collar bacon samples for NDMA. A Bonferroni analysis was performed on the samples showing significant differences ($P < 0.05$) by ANOVA. Comparisons for the Bonferroni analysis were between back and collar bacon for the following treatments: 1) immersion-cured bacon; control vs α -tocopherol 2) dry-salt cured bacon; control vs α -tocopherol 3) control dry-salt cured bacon vs control immersion-cured bacon; 4) α -tocopherol dry-salt cured bacon vs α -tocopherol immersion-cured bacon. Results reveal that NDMA levels in collar bacon were significantly different ($P < 0.05$) between immersion-cured bacon processed with and without α -tocopherol salts. Dry-cured collar bacon processed with and without α -tocopherol also differed significantly ($P < 0.05$) in NDMA levels.

Results of this study show that α -tocopherol effectively inhibits the formation of N-nitrosamines in Wiltshire bacon, especially with respect to NPYR. This agrees with the results of studies by Fiddler et al. (1978), Gray et al. (1982) and Reddy et al. (1982), who showed that NPYR was preferentially inhibited over NDMA by the use of α -tocopherol as a curing adjunct. The degree of inhibition of NPYR formation agrees also with the results reported by Skrypec et al. (1985) and Bernthal et al. (1986), who found the range of inhibition of NPYR formation to be 60-90% through the use of α -tocopherol-coated salts. These

researchers, however, studied the effects of α -tocopherol in belly bacon which had been cured by brine injection (Skrypec et al., 1985) or by the dry salt method (Bernthal et al., 1986).

There have been no reports on the effects of α -tocopherol-coated salts on the inhibition of N-nitrosamine formation in immersion-cured or dry salt-cured Wiltshire bacon. Results of this study indicate that α -tocopherol-coated salts are effective inhibitors of NPYR formation in Wiltshire bacon, irrespective of whether the bacon is produced by the traditional immersion curing process or by the dry salt procedure.

Effect of storage on N-nitrosamine levels in bacon. It is well established that the nitrite level in cured bacon influences the level of NPYR produced upon frying (Sen et al., 1974). Previously, it was believed that the initial level of nitrite in bacon dictated the quantities of N-nitrosamines formed during frying (Sen et al., 1974). More recently, however, it has been shown that the residual level of nitrite at the time of frying and not the initial level of nitrite is responsible for the formation of N-nitrosamines in fried bacon (Dudley, 1979; Sebranek, 1979).

As depicted in Tables 17 and 18, generally NPYR and NDMA concentrations were highest immediately after processing (Day 0). The high level of N-nitrosamines detected in fried bacon immediately after processing roughly correlates with high residual nitrite concentrations found at this time (Figures 3 - 6). Furthermore, as the residual nitrite concentrations decrease during storage, the levels of N-nitrosamines also decline (Tables 17 and 18). With few exceptions, the levels of the N-nitrosamines in this study decreased over time so

that the resultant levels of NPYR and NDMA were lower than those found initially.

In some instances, the levels of NPYR and NDMA in bacon after storage are slightly higher than the initial levels (i.e., dry cured control Wiltshire collar bacon, following 20 days of storage at 15°C (NDMA and NPYR); immersion cured α -tocopherol Wiltshire back and collar bacon, following 20 days of storage at 15°C (NPYR)). This increase in detectable N-nitrosamines during storage of the bacon has been found by other researchers and may be attributable to an increase in free amino acids and amines in the stored bacon (Pensabene et al., 1980). An increase of free proline (approximately 50%) has been reported in bacon stored at 2°C for 1 week (Lakritz et al., 1976; Gray and Collins, 1977).

N-Nitrosamines in the cooked-out fat of Wiltshire bacon. It is well established that during the frying of bacon, most of the N-nitrosamines formed are lost to the cooking vapors and only a portion of the N-nitrosamines remain in the fried bacon or cooked-out fat (Gough et al., 1976; Sen et al., 1976; Bharucha et al., 1979; Gray et al., 1982). Bharucha et al. (1979) found the concentration of volatile N-nitrosamines to be twice the concentration of those in the rasher. Other investigators have not found the levels of N-nitrosamines to vary as greatly between fried belly bacon and the cooked-out fat (Gray et al., 1982).

Gough et al. (1976) studied the distribution of N-nitrosamines in Wiltshire bacon and reported that cooked-out fat and cooking vapors contained a range of 0-30 and 10-160 ug/kg, respectively. In the present study, the cooked-out fat (drippings) were analyzed for the presence of both NPYR and NDMA (Table 22). The percent inhibition of

Table 22. N-Nitrosamine levels (ug/kg) in the cooked-out fat of Wiltshire bacon.

Treatment	<u>Control Samples</u>		<u>α-Tocopherol Samples</u>	
	NDMA	NPYR	NDMA	NPYR
<u>Immersion cured back bacon</u>				
Day 0	2.3 ^a	1.6 ^a	1.4	0.5
Day 20 5°C	1.2 ^a	1.4 ^a	1.2	3.4
Day 20 15°C	1.3 ^a	4.1 ^a	1.1	N.D.
<u>Immersion cured collar bacon</u>				
Day 0	2.9 ^a	2.6 ^a	0.9	N.D.
Day 20 5°C	1.4 ^a	1.8 ^a	N.D.	0.8
Day 20 15°C	1.7 ^a	4.9 ^a	0.6	N.D.
<u>Dry salted back bacon</u>				
Day 0	1.1	N.D.	1.5	3.4
Day 20 5°C	0.8	N.D.	0.3	N.D.
Day 20 15°C	N.D.	N.D.	0.3	N.D.
<u>Dry salted collar bacon</u>				
Day 0	1.3	N.D.	1.7	2.3
Day 20 5°C	N.D.	1.2	0.3	N.D.
Day 20 15°C	N.D.	N.D.	0.7	0.8

^aMean of 2 samples

^bN.D. = None detected

N-nitrosamines in the cooked-out fat through the use of α -tocopherol coated salts is illustrated in Table 23. The highest level of inhibition for NPYR was 68.8% and 69.0% for NDMA. Although the percent inhibition was somewhat lower than that found by Gray et al. (1982), different types of bacon systems were used for these studies. Nevertheless, it can be inferred from these data that α -tocopherol-coated curing salts assist in decreasing the quantities of N-nitrosamines formed in the cooked-out fat of Wiltshire bacon.

Table 23. Percent inhibition of N-nitrosamines in cooked-out fat using α -tocopherol-coated salts in Wiltshire bacon.

		<u>Immersion Cure</u>		<u>Dry Salt Cure</u>	
		NDMA	NPYR	NDMA	NPYR
<u>Backs</u>					
Day 0		39.1%	68.8%	NI ^a	NA ^b
Day 20	5°C	NI	NI	62.5%	NA
Day 20	15°C	15.4%	NA	NA	NA
<u>Collars</u>					
Day 0		69.0%	NA	NI	NA
Day 20	5°C	NA	55.6%	NA	NA
Day 20	15°C	64.7%	NA	NA	NA

^aNI = No inhibition of N-nitrosamines realized

^bNA = Not applicable

α -Tocopherol levels in Wiltshire cured bacon sides. Wiltshire bacon cured with α -tocopherol-coated salts were randomly selected and analyzed for residual α -tocopherol after 20 days of storage at 5 and 15°C. Results from these analyses are summarized in Tables 24 and 25. The mean values of residual α -tocopherol in immersion cured and dry salt cured Wiltshire bacon are 315 and 214 mg/kg, respectively. Gray

Table 24. Levels of α -tocopherol in immersion-cured Wiltshire bacon.

Sample Identification		mg/kg ^a	Standard Deviation	Range
Day 20 back	5°C	288.0	67.23	240.5-334.6
Day 20 collar	5°C	254.9	19.21	240.5-334.6
Day 20 collar	15°C	402.2	10.61	394.7-409.8
	mean	315.0		240.5-409.8

^aMean of 2 composite samples consisting of all 4 sides in treatment

Table 25. Levels of α -tocopherol in dry salt-cured Wiltshire bacon.

Sample Identification		mg/kg ^a	Standard Deviation	Range
Day 20 back	5°C	210.6	22.82	194.4-226.7
Day 20 collar	5°C	169.8	19.17	156.2-183.3
Day 20 collar	15°C	260.3	19.21	246.7-273.9
	mean	213.6		156.2-273.9

^aMean of 2 composite samples consisting of all 4 sides in treatment

et al. (1982) reported a residual α -tocopherol level of 360 mg/kg in dry cured belly bacon, while a range of residual α -tocopherol levels in pumped belly bacon of 232–313 mg/kg was reported by Fiddler et al. (1978). Both of these researchers applied an ingoing level of α -tocopherol of 500 mg/kg. The values obtained for residual α -tocopherol levels in this study are acceptable since variable processing procedures and bacon types were analyzed in this study and cannot be directly compared to bacon produced from pork bellies.

Stability of Wiltshire bacon during storage. Stability of the lipids in Wiltshire bacon prepared with and without α -tocopherol-coated salts was evaluated by the TBA method. TBA analyses were carried out over time on samples stored at 5 and 15°C (Tables 26 and 27). Since most bacon is not eaten immediately after processing, the statistical

Table 26. TBA^a values of Wiltshire bacon processed without α -tocopherol.

Treatment	Day of Storage					
	0	5	10	15	20	30
<u>Immersion cured</u>						
5°C back	0.14	0.23	0.40	0.32	0.10	0.01
5°C collar	0.08	0.26	0.35	0.23	0.17	0.19
15°C back	-	0.26	0.34	0.41	0.30	0.06
15°C collar	-	0.29	0.37	0.30	0.26	0.25
<u>Dry Salt cured</u>						
5°C back	0.32	0.20	0.25	0.61	0.39	0.33
5°C collar	0.35	0.29	0.23	0.21	0.25	0.24
15°C back	-	0.07	0.27	0.32	0.54	0.36
15°C collar	-	0.17	0.34	0.40	0.50	0.22

^aAnalyses were performed in duplicates consisting of a composite sample of all sides receiving the same treatment.

Table 27. TBA values of Wiltshire bacon processed with α -tocopherol.^a

Treatment	Day of Storage			
	0	5	15	20
<u>Immersion cured</u>				
5°C back	0.25	0.22	0.08	0.19
5°C collar	0.36	0.35	0.21	0.26
15°C back	-	0.23	0.28	0.24
15°C collar	-	0.34	0.20	0.35
<u>Dry Salt cured</u>				
5°C back	0.09	0.13	0.18	0.10
5°C collar	0.08	0.22	0.25	0.20
15°C back	-	0.38	0.30	0.20
15°C collar	-	0.26	0.57	0.19

^aAnalyses were performed in duplicates consisting of a composite sample of all sides receiving the same treatment.

analyses (ANOVA) were applied only to Day 15 data as this represents an average time which the bacon is in the distribution channel before being purchased by the consumer. Results of fatty acid analyses are listed in Appendices 4 and 5, and will not be discussed as the data are not critical to this study.

As shown in Tables 26 and 27, the TBA values obtained for immersion-cured and dry-cured Wiltshire bacon remain relatively constant throughout the storage period. Although sample variability may have caused slight deviations in the TBA values obtained, it appears that minimal lipid oxidation occurred during the storage period. Furthermore, the similarity between the TBA values in bacon cured with or without α -tocopherol indicates that the inclusion of α -tocopherol as a curing agent does not affect the degree of lipid oxidation during storage of Wiltshire bacon. A statistical evaluation

of back and collar bacon data by the ANOVA method indicates no significant differences ($P < 0.05$) between all treatments of collar bacon stored for 15 days at 5°C . A significant difference ($P < 0.05$) did exist, however, between back bacon from each treatment at Day 15 of 5°C storage.

The stability of stored bacon is related to the extent of lipid oxidation which has occurred in the unsaturated fatty acids of the meat. Changes in meat color, flavor and even nutritive value have been reported to occur as their fats are oxidized and interact with other meat constituents (Christopher et al., 1980). Peroxides are formed as an intermediate product during the oxidation of unsaturated fatty acids and are precursors to odor- and flavor-producing compounds (Buckley and Connolly, 1980). The inclusion of sodium nitrite in cured meats has been found to be responsible for the retardation of lipid oxidation (Cho and Bratzler, 1970).

Fooladi et al. (1979) reported that the addition of nitrite to beef and chicken resulted in a two-fold reduction in TBA values, while a five-fold reduction in TBA values occurred when nitrite was added to pork. The addition of nitrite to hams stored aerobically at 4°C has shown to significantly reduce ($P < 0.05$) lipid oxidation (MacDonald et al., 1980). Furthermore, it has been reported that canned, emulsified pork shoulders containing nitrite did not develop TBA values greater than 1.0 when stored for 14 weeks (Hadden et al., 1975). Researchers have demonstrated a correlation between TBA values and rancidity in pork products as observed by organoleptic evaluations (Turner et al., 1954; Younathan and Watts, 1959; Tarladgis et al., 1960). Generally, the TBA value at which rancidity became detectable was approximately

0.5. TBA values obtained in the present study agree with those of Buckley and Connolly (1980) and are well below that at which rancidity is observed in pork products.

The mechanism of action of nitrite in the inhibition of warmed-over flavor has not yet been completely resolved. Some researchers believe that nitrite reacts with iron porphyrins, forming a stable non-reactive compound, thus inhibiting the development of warmed-over flavors (Zipser et al., 1964; Igene et al., 1979; Igene et al., 1985). Igene et al. (1985) also reported that nitrite could inhibit WOF by stabilizing unsaturated lipids present in the membranes of meat tissues, or that nitrite could "tie-up" metal ions so that they are unavailable as catalysts of oxidative reactions.

Several meat additives have been researched in an attempt to decrease the extent of lipid oxidation in meats. Included in this list of compounds are butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), phosphate, ascorbate and citric acid, and sodium nitrite (Watts, 1950; Tims and Watts, 1958; MacDonald et al., 1980; Pearson and Gray, 1983).

Other researchers have attempted to increase "natural" antioxidants in meat prior to slaughter (Buckley and Connolly, 1980). These researchers reported that very little differences in TBA values existed between Wiltshire bacon which had been prepared from pigs fed vitamin E-enriched feed prior to slaughter or regular feed. The TBA values obtained from their study were below 0.20 after 14 weeks of storage at 5°C, and the authors suggested that nitrite may have been responsible for the unchanged oxidation levels owing to its antioxidant effect.

Sensory analysis of cooked Wiltshire bacon samples. On each taste panel day, both the collar and back portions of the pig side were analyzed by untrained panelists. Both the immersion cured and dry salt cured bacon were evaluated at the same sitting if the taste panel dates coincided. The panelists were asked to rank five criteria: flavor acceptability, color intensity, texture, saltiness and over-all acceptability (Appendix 1), in a manner similar to that described by Taylor et al. (1980). The panelists were not asked to evaluate which product they preferred, only to evaluate each sample individually. Raw data for the sensory analysis of Wiltshire bacon are presented in Appendices 6 - 9. As for the TBA analysis, only sensory data pertaining to the 15 day samples at 5°C will be discussed as this period of time is most representative of the time where consumers would purchase and eat the product.

A statistical analysis (ANOVA) of the sensory criterion of "flavor acceptability" revealed no significant differences ($P < 0.05$) between back bacon processed with or without α -tocopherol-coated salts, or between collar bacon processed in either of the above manners. A statistical difference ($P < 0.05$) in "overall acceptability" was shown to exist between back bacon cured with or without α -tocopherol as a curing adjunct. Collar bacon processed either with or without α -tocopherol-coated salts failed to indicate any significant differences for "overall acceptability." It appears that, in general, the panelists did not prefer one method of curing (i.e., dry-cure over immersion-cure) or that α -tocopherol interfered with the panelists' perception of bacon quality.

A comparison of the sensory data with those of Taylor et al. (1980) and Taylor et al. (1982) indicated that the salt levels traditionally used in the manufacture of Wiltshire bacon are much too high for consumers of this bacon in the United States. Since Wiltshire bacon is produced to contain 4 to 5% salt (Taylor et al., 1980), it is not surprising that panelists ranked the bacon "too salty" since bacon produced in the United States contains lower levels of salt (Kramlich et al., 1973). Other sensory assessments indicate that in general, back bacon was lighter in appearance and more moist than collar bacon. The ranking of flavor and overall evaluation appeared to be similar for back and collar bacon. Since this study evaluated whole rashers and not separate components of the rashers (fat and lean) as did previous studies, no further comparisons could be made with other studies (Taylor and Shaw, 1975; Taylor et al., 1980; Taylor et al., 1982).

Microbiology of Wiltshire bacon. Enumeration of the microorganisms associated with Wiltshire bacon is presented in Tables 28 - 30. Numbers of halophiles and organism growing on the Lactobacillus Agar were calculated for all treatments of the bacon in this study, while enumeration of the total plate counts and psychrotrophs were also performed on bacon sides cured without α -tocopherol. Individual values for total viable counts of bacteria on bacon sides after brine injection during processing and after the maturation period are listed in Appendices 10 - 13. Means of total viable counts of bacteria ($\log_{10}$) on the lean of immersion-cured and dry-cured bacon after the maturation period are 3.5 and 2.9. When bacon was processed with α -tocopherol-coated salts, the mean total viable counts were 2.6 and 3.2, respectively, for the immersion-cured

Table 28. Enumeration of bacteria on dry-cured Wiltshire bacon processed without α -tocopherol.^a

Treatment	Total Plate Count (log ₁₀)		Halophile Count (log ₁₀)		Psychrotroph Count (log ₁₀)		Lactobacillus Agar Count (log ₁₀)	
	5°C	15°C	5°C	15°C	5°C	15°C	5°C	15°C
<u>Back</u>								
Day 0		3.4					1.0	
Day 5	2.7	4.9	2.7	4.9	1.9	1.0	1.0	2.9
Day 10	3.6	6.7	3.7	6.7	3.1	4.1	1.0	4.2
Day 15	3.3	7.2	3.1	7.2	1.0	3.7	1.0	2.0
Day 20	3.8	7.6	3.9	7.4	1.0	4.8	1.4	1.0
<u>Collar</u>								
Day 0		3.8					1.0	
Day 5	3.5	6.2	3.5	6.2	1.0	3.8	1.0	1.0
Day 10	3.2	6.7	3.1	6.4	2.4	1.0	1.0	4.2
Day 15	3.3	7.1	3.3	7.3	1.0	2.9	1.0	2.0
Day 20	3.2	7.5	3.5	7.5	1.0	4.0	1.0	1.0

^aValues represent a numerical mean log₁₀ of duplicate plates in serial dilutions.

Table 29. Enumeration of bacteria on immersion-cured Wiltshire bacon processed without α -tocopherol.

Treatment	Total Plate Count ($\log_{10}$)		Halophile Count ($\log_{10}$)		Psychrotroph Count ($\log_{10}$)		Lactobacillus Agar Count ($\log_{10}$)	
	5°C	15°C	5°C	15°C	5°C	15°C	5°C	15°C
<u>Back</u>								
Day 0		4.0					1.0	
Day 5	3.6	6.3	3.3	4.0	1.2	1.0	1.0	1.4
Day 10	3.4	6.3	3.5	6.0	1.2	1.0	1.0	1.0
Day 15	3.0	7.0	3.6	6.9	1.0	1.0	1.0	1.0
Day 20	2.4	6.7	2.6	6.7	1.0	1.0	1.0	2.3
<u>Collar</u>								
Day 0		3.9		4.0		1.7	1.0	
Day 5	3.8	6.7	4.4	6.7	1.4	3.5	1.0	1.3
Day 10	4.0	6.2	3.7	6.1	1.0	4.6	1.0	1.0
Day 15	3.4	6.7	3.4	6.7	1.0	3.2	1.0	1.0
Day 20	3.6	6.7	2.7	6.7	1.0	1.0	1.0	1.0

^aValues represent a numerical mean $\log_{10}$ of duplicate plates in serial dilutions.

Table 30. Enumeration of bacteria on Wiltshire bacon processed with α -tocopherol.^a

Treatment	Dry-Cured				Immersion-Cured			
	Halophile Count ($\log_{10}$)		Lactobacillus Agar Count ($\log_{10}$)		Halophile Count ($\log_{10}$)		Lactobacillus Agar Count ($\log_{10}$)	
	5°C	15°C	5°C	15°C	5°C	15°C	5°C	15°C
<u>Back</u>								
Day 0		3.1		1.0	2.8		1.0	
Day 5	3.6	6.1	1.0	2.8	4.2	4.1	1.0	1.0
Day 15	5.3	6.9	3.6	6.5	4.5	5.6	1.0	1.0
Day 20	6.3	7.2	2.4	5.9	5.9	6.5	1.0	2.2
<u>Collar</u>								
Day 0		3.1		1.0	3.2		1.0	
Day 5	3.7	6.0	1.5	5.0	4.2	4.5	1.0	3.6
Day 15	5.2	6.8	1.6	5.1	4.1	6.0	2.1	1.0
Day 20	5.6	7.3	4.7	6.7	6.1	7.4	2.5	1.9

^aValues represent a numerical mean $\log_{10}$ of duplicate plates in serial dilutions.

and dry-cured bacon after the maturation period (Tables 31 and 32). Taylor et al. (1980) reported the mean of total viable counts to be 5.4 and 4.3 for the same sampling time for the immersion-cured and dry-cured bacon, respectively. Although the values for total viable counts in this study are low compared to those of a previous study (Taylor et al., 1980), this could be due to the processing technique followed in this study of rinsing the sides with water the evening before slicing. More importantly, the infrequent use of the processing equipment and facilities in the Meat Processing Laboratory may result in minimal buildup of microbial flora. This could then reflect in the lower contamination initially of the Wiltshire sides.

Stored bacon samples at 15°C generally contained greater microbial numbers than those bacon samples stored at 5°C. This was especially pronounced in total plate counts and halophilic organisms. This trend was also observed by Taylor et al. (1980, 1982). A comparison of the numbers of halophiles to those reported by Taylor and coworkers (1980, 1982) indicates that the number of bacteria are well within the expected range. The counts for organisms growing on Lactobacillus Agar, particularly in the bacon cured without α -tocopherol and also immersion-cured back bacon cured with α -tocopherol, are much lower in this study than those found by Taylor et al. (1980, 1982). This may be partially explained by the methods of sampling since the present study utilizes the whole rasher, whereas the above mentioned authors studied the lean portion of the bacon (separated from the fat). Furthermore, lactic acid bacteria are more common on the lean portions of Wiltshire bacon than the fat (Taylor and Shaw, 1975).

Table 31. Total viable counts of microorganisms during processing of immersion-cured Wiltshire bacon.^a

Treatment	Sample	Mean Total Viable ^b Count/g(log ₁₀)
<u>After Brine Injection</u>		
Without α -tocopherol	rind	4.3
Without α -tocopherol	pleura	3.9
With α -tocopherol	rind	3.8
With α -tocopherol	pleura	3.8
<u>After Immersion Period</u>		
Without α -tocopherol	rind	4.4
Without α -tocopherol	pleura	4.2
With α -tocopherol	rind	3.5
With α -tocopherol	pleura	3.8
<u>After Maturation Period</u>		
Without α -tocopherol	rind	4.0
Without α -tocopherol	pleura	3.5
With α -tocopherol	rind	3.0
With α -tocopherol	pleura	2.6

^aValues represent averages of 4 sides per treatment.

^bValues represent a numerical mean log₁₀ of duplicate plates in serial dilutions.

Table 32. Total viable counts of microorganisms during processing of dry-cured Wiltshire bacon.^a

Treatment	Sample	Mean Total Viable ^b Count/g(log ₁₀)
<u>After Brine Injection</u>		
Without α -tocopherol	rind	4.3
Without α -tocopherol	pleura	3.9
With α -tocopherol	rind	3.4
With α -tocopherol	pleura	3.2
<u>After Maturation Period</u>		
Without α -tocopherol	rind	3.5
Without α -tocopherol	pleura	2.9
With α -tocopherol	rind	2.8
With α -tocopherol	pleura	3.2

^aValues represent averages of 4 sides per treatment.

^bValues represent a numerical mean log₁₀ of duplicate plates in serial dilutions.

Generally, the microbial population on back and collar bacon samples of the same treatment do not appear to differ greatly. Additionally, the total plate counts of bacon cured without α -tocopherol were similar to the population of microorganisms found on their α -tocopherol-cured counterparts. Taylor et al. (1980) reported that dry-cured bacon initially contained fewer bacteria than immersion-cured bacon; however, an increase in the number of bacteria on the dry-cured bacon during storage occurred until the number of microorganisms present was greater than that on the immersion-cured bacon sides. It appears that these authors were correct in their assumption that increased application of curing salts in the back region of the carcass during processing would have a negative effect on the resulting numbers of bacteria growing on this region.

SUMMARY AND CONCLUSIONS

The usage of α -tocopherol as a curing adjunct in Wiltshire bacon was examined. Immersion-cured and dry-salt-cured bacon samples were evaluated for N-nitrosamine content and the extent of inhibition of N-nitrosamines in bacon samples cured with α -tocopherol. The effects of α -tocopherol on TBA and sensory values were also explored. Additionally, variability of microbial numbers on Wiltshire bacon sides in α -tocopherol-treated and untreated sides were investigated.

As a result of this study, several inferences can be made about Wiltshire bacon processed with α -tocopherol.

First, the inclusion of α -tocopherol in the curing ingredients of Wiltshire sides markedly reduced the levels of N-nitrosamines formed upon frying. This is evident by inhibition levels as great as 86%. α -Tocopherol had no effect on the degree of lipid oxidation as indicated by the relatively constant TBA values during storage. These results were expected since sodium nitrite present in the cure also protects against oxidation. Although the overall perception of the bacon samples by the panelists was "too salty," α -tocopherol did not appear to influence the overall flavor of the product. Finally, a microbiological analysis of the sides during processing indicates that the number of bacteria was not influenced by the presence of α -tocopherol. While the Lactobacillus Agar bacterial counts on some treatment samples appeared lower than expected, the counts of

halophilic organisms were in agreement with previous researchers (Taylor et al., 1980 and 1982). These results indicate that the addition of α -tocopherol may be a beneficial factor in the production of Wiltshire bacon.

The inclusion of α -tocopherol substantially reduces the N-nitrosamines formed in the bacon, does not affect the stability of the product and does not appear to be perceptible by taste panelists. Furthermore, Wiltshire bacon has similar microbiological characteristics regardless of whether the bacon contains α -tocopherol. The inclusion of α -tocopherol in the curing ingredients is a viable adjunct to immersion-cured and dry-salt-cured Wiltshire bacon sides.

BIBLIOGRAPHY

BIBLIOGRAPHY

- Ala-Huikko, K., Nurmi, E., Pajulahti, H., and Raevuori, M. 1977. Effect of nitrite, storage temperature and time on Clostridium botulinum type A toxin formation in liver sausage. Eur. J. Appl. Microbiol. 4:145.
- American Meat Institute, 1944. Pork Operations. Institute of Meat Packing, Univ. of Chicago, Chicago, IL.
- A.O.A.C. 1970. Official Methods of Analysis, 11th Edition, Association of Official Analytical Chemists, Washington, D.C.
- A.O.A.C. 1984. Official Methods of Analysis, 14th Edition, Association of Official Analytical Chemists, Washington, D.C.
- American Public Health Association, 1972. Standard Methods for the Examination of Dairy Products, 13th Edition, New York.
- Bacus, J.N. 1979. Reduces nitrosamines. Food Eng. 51:24.
- Barbe, C.D., Mandigo, R.W. and Henrickson, R.L. 1966. Bacterial flora associated with rapid-processed ham. J. Food Sci. 31:988.
- Barbe, C.D. and Henrickson, R.L. 1967. Bacteriology of rapid cured ham. Food Technol. 21(8):103.
- Benedict, R.C. 1980. Biochemical basis for nitrite-inhibition of Clostridium botulinum in cured meat. J. Food Protect. 43:877.
- Bernthal, P.H., Gray, J.I., Mandagere, A.K., Ikins, W.G., Cuppett, S.L., Booren, A.M., and Price, J.F. 1986. Use of antioxidant-coated salts as N-nitrosamine inhibitors in dry- and brine-cured bacon. J. Food Protect. 19:58.
- Bharucha, K.R., Cross, C.K., and Rubin, L.J. 1979. Mechanism of N-nitrosopyrrolidine formation in bacon. J. Agric. Food Chem. 27:63.
- Bharucha, K.R., Cross, C.K., and Rubin, L.J. 1980. Long-chain acetals of ascorbic and erythorbic acids as anti-nitrosamine agents for bacon. J. Agric. Food Chem. 28:1274.
- Buckley, J. and Connolly, J.F. 1980. Influence of alpha-tocopherol (Vitamin E) on storage stability of raw pork and bacon. J. Food Protect. 43:265.

- Cassens, R.G., Sebranek, J.G., Kubberod, G., and Woolford, G. 1974. Where does the nitrite go? Food Prod. Dev., Dec.
- Cassens, R.G., Greaser, M.L., Ito, T., and Lee, M. 1979. Reactions of nitrite in meat. Food Technol 31(7):46.
- Cervený, J.G. 1980. Effects of changes in the production and marketing of cured meats on the risk of botulism. Food Technol. 34(5):240.
- 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 botulinal 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 nitrosamine formation in perishable canned comminuted cured meat. Appl. Microbiol. 25:357.
- 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. Microbiol. 27:733.
- Christiansen, L.N., Tompkin, R.B., Shaparis, A.B. 1975. Effect of sodium nitrite and nitrate on Clostridium botulinum growth and toxin production in a summer style sausage. J. Food Sci. 40:488.
- Christopher, F.M., Carpenter, Z.L., Dill, C.W., Smith, G.C., and Vanderzant, C. 1980. Microbiology of beef, pork and lamb stored in vacuum or modified gas atmospheres. J. Food Protect. 43:259.
- Cook, W.H. and White, W.H. 1940. Canadian Wiltshire bacon. II. Chloride, nitrate, and nitrite content of bacon and pickle. Can. J. Res, D. 18:135.
- Cornish, D.G. and Mandigo, R.W. 1974. Accelerated pork processing: A quantitative study of bacterial flora of cured and smoked hams. J. Food Sci. 39:605.
- Crosby, N.T., Foreman, J.K., Palframan, J.F., and Sawyer, R. 1972. Estimation of steam-volatile N-nitrosamines in foods at the 1 ug/kg level. Nature. 238:342.
- Crosby, N.T. and Sawyer, R. 1976. N-Nitrosamines: A review of chemical and biological properties and their estimation in foodstuffs. Adv. Food Res. 22:1.

- Cuthbertson, A. 1982. Hot processing in Britain. Proc. Inter. Symp. Meat Sci. and Technol. in cooperation with National Live Stock and Meat Board. Pub. National Live Stock and Meat Board, Chicago, IL, Univ. of Nebraska, Lincoln, NE.
- Dudley, R. 1979. How to keep your bacon out of trouble. *Meat Ind.* 26(2):24.
- Ender, F., Harve, G., Helgebostad, A., Koppang, N., Madsen, R., and Ceh, L. 1964. Isolation and identification of a hepatotoxic factor in herring meal produced from sodium nitrite preserved herring. *Naturwissenschaften.* 51:637.
- Federal Register, 1975. 40, 52614.
- Federal Register, 1985. 49, 13346.
- Fiddler, W., Pensabene, J.W., Piotrowski, E.G., Doerr, R.C., and Wasserman, A.E. 1973. Use of sodium ascorbate or erythorbate to inhibit formation of N-nitrosodimethylamine in frankfurters. *J. Food Sci.* 38:1084.
- Fiddler, W., Pensabene, J.W., Fagan, J.C., Thorne, E.J., Piotrowski, E.G., and Wasserman, A.E. 1974. The role of lean and adipose tissue on the formation of nitrosopyrrolidine in fried bacon. *J. Food Sci.* 39:1070.
- 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 nitrosamines in fried bacon by the use of cure-solubilized α -tocopherol. *J. Agric. Food Chem.* 26:653.
- Fooladi, M.H., Pearson, A.M., Coleman, T.H., and Merkel, R.A. 1979. The role of nitrite in preventing development of warmed-over flavor. *Food Chem.* 4:283.
- Gardner, G.A. 1971. Microbiological and chemical changes in lean Wiltshire bacon during aerobic storage. *J. Appl. Bact.* 34(3):645.
- Gardner, G.A. 1975. The enumeration of microorganisms on Wiltshire bacon sides: A scheme for use in quality control. *J. Fd. Technol.* 10:181.
- Gardner, G.A. and Patton, J. 1969. Variations in the composition of the flora on a Wiltshire cured bacon side. *J. Fd. Technol.* 4:125.
- Garrard, E.H. and Lochhead, A.G. 1939. A study of bacteria contaminating sides for Wiltshire bacon with special consideration of their behavior in concentrated salt solutions. *Can. J. Res.* 17:45.

- Gibbons, N.E. 1940a. Canadian Wiltshire bacon. V. Quantitative bacteriological studies on curing pickles. Can. J. Res, D. 18:191.
- Gibbons, N.E. 1940b. Canadian Wiltshire bacon. VI. Quantitative bacteriological studies on product. Can. J. Res, D. 18:202.
- Gill, J.L. 1978. Design and Analysis of Experiments in the Animal and Medical Sciences. Iowa State Univ. Press. Ames, Iowa.
- Gough, T.A., Goodhead, K., and Walters, C.L. 1976. Distribution of some volatile nitrosamines in cooked bacon. J. Sci. Fd. Agric. 27:181.
- Gray, J.I. 1976. N-Nitrosamines and their precursors in bacon: A review. J. Milk Food Technol. 39:686.
- Gray, J.I. and Collins, M.E. 1977. A comparison of proline and putrescine as precursors of N-nitrosopyrrolidine in nitrite-treated pork systems. J. Food Sci. 42:1034.
- Gray, J.I., Collins, M.E., and Russell, L.F. 1977. Formation of N-nitrosohydroxypyrrolidine in model and cured meat systems. Can. Inst. Food Sci. Technol. J. 10:36.
- Gray, J.I., Collins, M.E., and MacDonald, B. 1978. Precursors of dimethylnitrosamines in fried bacon. J. Food Prot. 41:31.
- Gray, J.I. and Randall, C.J. 1979. The nitrite/N-nitrosamine problem in meats: An update. J. Food Protect. 42:1034.
- Gray, J.I. 1981. Formation of N-nitroso compounds in foods. In N-Nitroso Compounds, ACS Symp. Series No. 174, p. 165. Amer. Chem. Soc., Washington, D.C.
- Gray, J.I., Reddy, S.K., Price, J.F., Mandagere, A., and Wilkens, W.F. 1982. Inhibition of N-nitrosamines in bacon. Food Technol. 36(6):39.
- Greenberg, R.A., Silliker, J.H., and Fatta, L.D. 1958. The influence of sodium chloride on toxin production and organoleptic breakdown in perishable cured meat inoculated with Clostridium botulinum. Food Technol. 13:509.
- Greenberg, R.A. 1973. Ascorbate and nitrosamine formation in cured meats. Proc. Int. Symp. Nitrite Meat Prod., Zeist, Pudoc, Wageningen, p.179.
- Hadden, J.P., Ockerman, H.W., Cahill, V.R., Parrett, N.A., and Borton, R.J. 1975. Influence of sodium nitrite on the chemical and organoleptic properties of comminuted pork. J. Food Sci. 40:626.
- Havery, D.C. and Fazio, T. 1985. Human exposure to nitrosamines from foods. Food Technol. 39(1):80.

- Henrickson, R.L. and Smith, R.E. 1967. Effect of rapid processing on the properties of freshly slaughtered pork. *Trans. Amer. Soc. Agric. Eng.* 10:185.
- Holley, R.A. 1981. Review of the potential hazard from botulism in cured meats. *Can. Inst. Food Sci. Technol. J.* 14:183.
- Hotchkiss, J.H. and Vecchio, A.J. 1985. Nitrosamines in fried-out bacon fat and its use as a cooking oil. *Food Technol.* 39(1):67.
- Hustad, G.O., Cervený, 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 nitrosamine formation in wieners. *Appl. Microbiol.* 26:22.
- Hwang, L.S. and Rosen, J.D. 1976. Nitrosopyrrolidine formation in fried bacon. *J. Agric. Food Chem.* 24:1152.
- Igene, J.O., King, J.A., Pearson, A.M., and Gray, J.I. 1979. Influence of heme pigments, nitrite and non-heme iron on development of warmed-over flavor (WOF) in cooked meat. *J. Agric. Food Chem.* 27:838.
- Igene, J.O., Yamauchi, K., Pearson, A.M., Gray, J.I., and Aust, S.D. 1985. Mechanisms by which nitrite inhibits the development of warmed-over flavor (WOF) in cured meat. *Food Chem.* 18:1.
- Ingram, M. 1952. Internal bacteria taints ("bone taint" or "souring") of cured pork legs. *J. Hyg., Lond.* 50:165.
- Ingram, M. 1960. Bacterial multiplication in packed Wiltshire bacon. *J. Appl. Bact.* 23:206.
- Ingram, M. 1973. The microbiological effects on nitrite. In Proc. 2nd Intl. Symp. Nitrite Meat Prod., Zeist, Pudoc, Wageningen, p. 63.
- Ingram, M. and Hobbs, B.C. 1954. The bacteriology of "pasteurized" canned hams. *J. Roy. Sanit. Inst.* 74:1151.
- Jespersen, N.J. and Riemann, H. 1958. The microbiology of fish and meat curing brines. In 2nd Intl. Symp. on Food Micro., Cambridge, London, p 177. H.M.S.O.
- Jolley, P.D. 1979. The accumulation of unacceptable high levels of nitrite in vacuum packed back bacon. *J. Fd. Technol.* 14:81.
- Kastner, C.L. 1982. Hot processing-overview. Proc. Inter. Symp. Meat Sci. and Technol. in cooperation with National Live Stock and Meat Board. Pub. National Live Stock and Meat Board, Chicago, IL. Univ. of Nebraska, Lincoln, NE.

- Kerr, R.H., Marsh, C.T.N., Schroeder, W.F., and Boyer, E.A. 1926. The use of sodium nitrite in curing meat. *J. Agric. Res.* 33:541.
- Kotula, A.W. 1981. Microbiology of hot-boned and electrostimulated meat. *J. Food Protect.* 44:545.
- Kramlich, W.E., Pearson, A.M., and Tauber, F.W. 1973. Processed Meats. AVI Pub. Co. Inc. Westport, CT.
- Lakritz, L., Spinelli, A.M., and Wasserman, A.E. 1976. Effect of storage on the concentration of proline and other free amino acids in pork bellies. *J. Food Sci.* 41:879.
- Lee, C. and Cassens, R.G. 1980. Effect of sodium chloride on residual nitrite. *J. Food Sci.* 45:267.
- Lee, M.L., Gray, J.I., Pearson, A.M., and Kakuda, Y. 1983. Formation of N-nitrosopyrrolidine in fried bacon: Model system studies. *J. Food Sci.* 48:820.
- Leistner, L., Rodel, W., and Krespien, K. 1981. Microbiology of meat and meat products in high- and intermediate-moisture ranges. In: Water Activity: Influences in Food Quality. Rockland, L.B. and Stewart, G.F. (eds.) Academic Press, London.
- MacDonald, B., Gray, J.I., Kakuda, Y., and Lee, M.L. 1980. Role of nitrite in cured meat flavor: chemical analysis. *J. Food Sci.* 45:899.
- Magee, P.N. and Barnes, J.M. 1956. The production of malignant primary hepatic tumors in the rat by feeding dimethylnitrosamine. *Brit. J. Cancer.* 10:114.
- Mandigo, R.W. and Henrickson, R.L. 1966. Influence of hot-processing pork carcasses on cured ham. *Food Technol.* 20(4):186.
- Mergens, W.J. and Newmark, H.L. 1979. The use of alpha-tocopherol in bacon processing: An update. Proc. Meat Industry Research Conf., American Meat Institute Foundation, Chicago, IL, p. 79.
- Mirvish, S.S. 1970. Kinetics of dimethylamine nitrosation in relation to nitrosamine carcinogenesis. *J. Natl. Cancer Inst.* 44:633.
- Morrison, W.R. and Smith, L.M. 1964. Preparation of fatty acid methyl esters and dimethylacetals from lipids with boron fluoride-methanol. *J. Lipid Res.* 5:600.
- 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-nitrosodimethylamine in cured pork containing added dimethylamine. *J. Sci. Fd. Agric.* 26:47.

- 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., Edwards, R.A., and Gough, T.A. 1977. The preferential formation of volatile N-nitrosamines in the fat of fried bacon. *J. Sci. Fd. Agric.* 28:1025.
- 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.
- National Academy of Sciences. 1981. The Health Effects of Nitrate, Nitrite and Nitroso Compounds. National Academy Press, Washington, D.C.
- Nitrite Safety Council. 1980. A survey of 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. Inst. Food Technol. J.* 2:79.
- Owens, J.L. and Kinast, O.E. 1980. An improved procedure for the determination of volatile N-nitrosamines in bacon grease by using the mineral oil distillation-thermal energy analyzer method. *J. Agric. Food Chem.* 28:1262.
- Patterson, R.L.S., Taylor, A.A., Mottram, D.S., and Gough, T.A. 1976. Localized occurrence of N-nitrosopyrrolidine in fried bacon. *J. Sci. Fd. Agric.* 27:257.
- Pearson, A.M. and Gray, J.I. 1983. Mechanism responsible for warmed-over flavor in cooked meat. In: The Maillard Reaction in Foods and Nutrition. Eds., G.R. Waller and M.S. Feather. *Am. Chem. Soc. Symp. Series* 215:287.
- Pensabene, J.W., Fiddler, W., Gates, R.A., Fagan, J.C., and Wasserman, A.E. 1974. Effect of frying and other cooking conditions on 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 formulations on the inhibition of nitrosopyrrolidine formation in model systems. *J. Food Sci.* 43:801.
- Pensabene, J.W., Feinberg, J.I., Dooley, C.J., Phillips, J.G., and Fiddler, W. 1979a. Effect of pork belly composition and nitrite level on nitrosamine formation in fried bacon. *J. Agric. Food Chem.* 27:842.
- Pensabene, J.W., Feinberg, J.I., Piotrowski, E.G., and Fiddler, W. 1979b. Occurrence and determination of N-nitrosoproline and N-nitrosopyrrolidine in cured meat products. *J. Food Sci.* 44:1700.

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100-100000-0000

- Pensabene, J.W., Fiddler, W., Miller, A.J., and Phillips, J.G. 1980. Effect of preprocessing procedures for green bellies on N-nitrosopyrrolidine formation in bacon. J. Agric. Food Chem. 28:966.
- Pierson, M.D. 1978. The evaluation of potassium sorbate as an inhibitor of Clostridium botulinum growth and toxin production in bacon. Report of March 14 to Monsanto Co., St. Louis, MO.
- Pivnick, H., Johnston, M.A., Thacker, C., and Loynes, R. 1970. Effect of nitrite on destruction and germination of Clostridium botulinum and putrefactive anaerobes 3679 and 3679h in meat and in buffer. Can. Inst. Food Technol. J. 3:103.
- Pulliam, J.D. and Kelly, D.C. 1965. Bacteriological comparisons of hot processed and normally processed hams. J. Milk Food Technol. 28:285.
- Reddy, S.K., Gray, J.I., Price, J.F., and Wilkens, W.F. 1982. Inhibition of N-nitrosopyrrolidine in dry cured bacon by α -tocopherol-coated salt systems. J. Food Sci. 47:1598.
- Roberts, T.A. and Smart, J.L. 1976. The occurrence and growth of Clostridium sps. in vacuum-packed bacon with particular reference to C. perfringens (welchii) and C. botulinum. J. Fd. Technol. 11:229.
- Scanlan, R.A. and Reyes, F.G. 1985. An update on analytical techniques for N-nitrosamines. Food Technol. 39(1):95.
- Sebranek, J.G. 1979. Advances in the technology of nitrite use and consideration of alternatives. Food Technol. 33(7):58.
- Sen, N.R. 1980. Nitrosamines. In: Safety of Foods. Graham, H.D. (ed.) AVI Publishing Co., Westport, CT.
- Sen, N.P., Donaldson, B., Iyengar, J.R., and Panalaks, T. 1973. Nitrosopyrrolidine and dimethylnitrosamine in bacon. Nature. 241:473.
- Sen, N.P., Iyengar, J.R., Donaldson, B.A., Panalaks, T. 1974. Effect of sodium nitrite concentration on the formation of nitrosopyrrolidine and dimethylnitrosamine in fried bacon. J. Agric. Food Chem. 22:540.
- Sen, N.P., Donaldson, B., Seaman, S., Iyengar, J.R., and Miles, W.F. 1976. Inhibition of nitrosamine formation in fried bacon by propyl gallate and L-ascorbyl palmitate. J. Agric. Food Chem. 24:397.
- Sen, N.P., Seaman, S., and Miles, W.F. 1976. Dimethylnitrosamine and nitrosopyrrolidine in fumes produced during the frying of bacon. Food Cosmet. Toxicol. 14:167.

- Sen, N.P., Seaman, S., and Miles, W.F. 1979. Volatile nitrosamines in various cured meat products: Effect of cooking and recent trends. *J. Agric. Food Chem.* 27:1352.
- Skrypec, D.J., Gray, J.I., Mandagere, A.K., Booren, A.M., Pearson, A.M., and Cuppett, S.L. 1985. Effect of bacon composition and processing on N-nitrosamine formation. *Food Technol.* 39(1):74.
- Statutory Instruments. 1971. Food and Drugs. Composition and Labelling. S.I. No. 882, The Preservatives in Food (Amendment) Regulations, 1971.
- Tanaka, N. 1980. Effect of d- α -tocopherol on antibotulinal effectiveness of sodium nitrite in bacon. *J. Food Sci.* 47:1797.
- Tanaka, N., Gordon, N.M., Lindsay, R.C., Meske, L.M., Doyle, M.P., and Traisman, E. 1985. Sensory characteristics of reduced nitrite bacon manufactured by the Wisconsin Process. *J. Food Protect.* 48:687.
- Tarladgis, B.G., Watts, B.M., Younathan, M.T., and Dugan, L.R., Jr. 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. *J. Amer. Oil Chem. Soc.* 37:44.
- Taylor, A.A. and Shaw, B.G. 1975. Wiltshire curing with and without nitrate. I. Vacuum packed sliced back bacon. *J. Fd. Technol.* 10:157.
- Taylor, A.A., Shaw, B.G., and Jolley, P.D. 1976. Wiltshire curing with and without nitrate. II. Vacuum packed collar bacon and vacuum packed bacon from pigs with high ultimate pH. *J. Fd. Technol.* 11:589.
- Taylor, A.A., Shaw, B.G., and Jolley, P.D. 1980. A modern dry-salting process for Wiltshire bacon. *J. Fd. Technol.* 15:301.
- Taylor, A.A., Shaw, B.G., Jolley, P.D., and Nute, G.R. 1982. Hot curing Wiltshire bacon. *J. Fd. Technol.* 17:339.
- Thompson, J.N. and Hatina, G. 1979. Determination of tocopherols and tocotrienols in foods and tissues by high performance liquid chromatography. *J. Liquid Chromatog.* 2:327.
- Tims, M.J. and Watts, B.M. 1958. Protection of cooked meats with phosphates. *Food Technol.* 12:240.
- 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. 1980. Botulism from meat and poultry products - a historical perspective. *Food Technol.* 34(5):229.

- Troller, J.A. and Christian, J.H.B. 1978. Water Activity and Food. Academic Press, N.Y.
- Turner, E.W., Paynter, W.D., Montie, E.J., Bessert, M.W., Struck, G.J., and Olson, F.C. 1954. Use of 2-thiobarbituric acid reagent to measure rancidity in frozen pork. Food Technol. 8:326.
- Walters, C.L., Edwards, M.W., Elsey, T.S., and Martin, M. 1976. The effect of antioxidants on the production of volatile nitrosamines during the frying of bacon. Z. Lebensm. Unters-Forsch. 162:377.
- Warthesen, J.J., Bills, D.D., Scanlan, R.A., and Libbey, L.M. 1976. N-Nitrosopyrrolidine collected as a volatile during heat-induced formation in nitrite-containing pork. J. Agric. Food Chem. 24:892.
- Watts, B.M. 1950. Polyphosphates as synergistic antioxidants. J. Am. Oil Chem. Soc. 27:48.
- Younathan, M.T. and Watts, B.M. 1959. Relationship of meat pigments to lipid oxidation. Food Res. 24:728.
- Zipser, M.W. and Watts, B.M. 1962. A modified 2-thiobarbituric acid (TBA) method for determination of malonaldehyde in cured meats. Food Technol. 16(7):102.
- Zipser, M.W., Kwon, T.W., and Watts, B.M. 1964. Oxidative changes in cured frozen cooked pork. J. Agric. Food Chem. 12:105.

APPENDICES

Appendix 1

SENSORY EVALUATION OF WILTSHIRE BACON

NAME _____ SAMPLE # _____ DATE _____

<u>Flavor Acceptability</u>	<u>Color Intensity</u>	<u>Mouth Feel</u>	<u>Saltiness</u>	<u>Overall Acceptability</u>
Like Very much	Very Dark	Very Juicy	Very Salty	Very Desirable
Like Moderately	Moderately Dark	Moderately Juicy	Moderately Salty	Moderately Desirable
Like Slightly	Slightly Dark	Slightly Juicy	Slightly Salty	Slightly Desirable
Neither like Nor dislike	Neither dark Nor light	Neutral	Neutral	Neutral
Dislike Slightly	Slightly Light	Slightly Dry	Slightly Under Salty	Slightly Undesirable
Dislike Moderately	Moderately Light	Moderately Dry	Moderately Under Salty	Moderately Undesirable
Dislike Very much	Very Light	Very Dry	Very Under Salty	Very Undesirable

COMMENTS: _____

This is not a comparison - only an evaluation for an individual product

Appendix 2. Residual nitrite concentration in Wiltshire bacon processed without α -tocopherol.

		(mg/kg)				
		Day 0	Day 5	Day 10	Day 15	Day 20
<u>IMMERSION CURE</u>						
Back	5°C	105.9±6.3	86.4±7.0	65.3±16.2	60.6±15.2	42.9±13.6
Collar	5°C	93.9±9.7	103.1±12.2	76.9±8.4	71.7±8.0	54.4±25.6
Back	15°C	105.9±6.3	49.7±2.4	46.4±8.8	34.6±7.0	24.8±5.7
Collar	15°C	93.9±9.7	72.7±28.9	62.9±19.2	34.6±1.1	37.1±6.1
<u>DRY CURE</u>						
Back	5°C	55.7±16.4	67.5±8.1	45.0±3.4	40.2±4.5	26.9±4.7
Collar	5°C	67.1±34.5	96.3±18.4	51.6±19.9	66.9±46.0	59.5±20.3
Back	15°C	55.7±16.4	52.5±3.4	29.1±1.9	28.2±0.8	16.7±1.8
Collar	15°C	67.1±34.5	64.9±21.6	29.1±17.4	31.9±6.2	29.6±2.9

^aMean of duplicate samples for each side in treatment.

Appendix 3. Residual nitrite concentration in Wiltshire bacon processed with α -tocopherol.

		(mg/kg)			
		Day 0	Day 5	Day 15	Day 20
		<u>IMMERSION CURE</u>			
Back	5°C	133.9 $\pm$ 8.9	60.2 $\pm$ 17.0	49.5 $\pm$ 13.1	33.8 $\pm$ 10.7
Collar	5°C	129.0 $\pm$ 50	51.5 $\pm$ 21.5	34.8 $\pm$ 8.5	45.0 $\pm$ 18.1
Back	15°C	133.9 $\pm$ 8.9	47.4 $\pm$ 14.4	50.2 $\pm$ 26.7	24.9 $\pm$ 9.2
Collar	15°C	129.0 $\pm$ 50.1	40.1 $\pm$ 18.4	33.7 $\pm$ 25.5	17.6 $\pm$ 8.8
		<u>DRY CURE</u>			
Back	5°C	53.0 $\pm$ 8.1	23.6 $\pm$ 2.1	10.2 $\pm$ 0.9	8.0 $\pm$ 2.2
Collar	5°C	69.7 $\pm$ 7.5	31.1 $\pm$ 2.4	13.9 $\pm$ 3.0	9.7 $\pm$ 5.2
Back	15°C	53.0 $\pm$ 8.1	19.5 $\pm$ 1.8	10.2 $\pm$ 1.8	4.4 $\pm$ 0.0
Collar	15°C	69.7 $\pm$ 7.5	19.5 $\pm$ 1.8	14.1 $\pm$ 7.0	10.7 $\pm$ 5.7

^aMean of duplicate samples for each side in treatment.

Appendix 4. Fatty acid composition of Wiltshire bacon cured without α -tocopherol.

Bacon samples	C14	C16	C16:1	C18	C18:1	C18:2
<u>IMMERSION CURE</u>						
(%)						
Day 0 back	1.3 $\pm$ 0.21	24.1 $\pm$ 0.64	2.5 $\pm$ 0.14	14.3 $\pm$ 0.71	45.5 $\pm$ 0.42	10.1 $\pm$ 0.64
Day 0 collar	1.2 $\pm$ 0.07	22.7 $\pm$ 2.33	2.2 $\pm$ 0.14	14.2 $\pm$ 2.12	41.1 $\pm$ 4.88	16.4 $\pm$ 9.62
Day 20 5°C back	1.3 $\pm$ 0.14	22.9 $\pm$ 0.21	3.1 $\pm$ 0.14	10.8 $\pm$ 0.57	47.0 $\pm$ 1.56	11.4 $\pm$ 0.71
Day 20 5°C collar	1.3 $\pm$ 0.00	23.0 $\pm$ 0.14	3.1 $\pm$ 0.42	10.9 $\pm$ 1.06	46.6 $\pm$ 1.77	12.1 $\pm$ 0.71
Day 20 15°C back	1.4 $\pm$ 0.07	24.0 $\pm$ 0.07	2.9 $\pm$ 0.49	12.3 $\pm$ 1.20	47.7 $\pm$ 1.56	11.0 $\pm$ 0.85
Day 20 15°C collar	1.3 $\pm$ 0.07	23.0 $\pm$ 0.00	3.7 $\pm$ 0.07	9.8 $\pm$ 0.21	47.8 $\pm$ 1.13	12.6 $\pm$ 0.28
<u>DRY SALT CURE</u>						
Day 0 back	1.4 $\pm$ 0.07	24.8 $\pm$ 2.76	2.3 $\pm$ 1.13	12.7 $\pm$ 1.63	53.5 $\pm$ 4.38	10.9 ^b
Day 0 collar	<1.0 ^c	23.2 $\pm$ 1.70	2.7 $\pm$ 0.21	13.9 $\pm$ 0.28	49.7 $\pm$ 3.11	10.6 $\pm$ 0.92
Day 20 5°C back	1.2 ^d	23.3 $\pm$ 0.28	2.6 $\pm$ 0.21	14.6 $\pm$ 0.28	47.1 $\pm$ 1.48	12.0 $\pm$ 2.05
Day 20 5°C collar	1.3 $\pm$ 0.07	23.3 $\pm$ 0.64	2.5 $\pm$ 0.14	14.6 $\pm$ 1.13	45.5 $\pm$ 0.14	12.0 $\pm$ 1.70
Day 20 15°C back	1.3 $\pm$ 0.07	24.4 $\pm$ 1.34	2.7 $\pm$ 0.00	14.0 $\pm$ 0.35	46.5 $\pm$ 1.77	9.7 $\pm$ 0.78
Day 20 15°C collar	1.3 $\pm$ 0.00	24.1 $\pm$ 0.71	2.9 $\pm$ 0.14	13.4 $\pm$ 0.92	45.0 $\pm$ 0.35	11.1 $\pm$ 0.78

^aAverages of 2 samples representative of the treatment and sampling period

^bValue represented by 1 sample - peak was not integrated for second sample

^cArea of both peaks were <1% of total area

^dArea of peak of 1 sample was <1% of total area

Appendix 5. Fatty acid composition of Wiltshire bacon cured with α -tocopherol.

Bacon samples	C14	C16	C16:1	C18	C18:1	C18:2
<u>IMMERSION CURE</u>						
(%)						
Day 0 back	1.4 $\pm$ 0.07	23.5 $\pm$ 1.63	3.2 $\pm$ 0.42	12.3 $\pm$ 0.71	46.3 $\pm$ 0.78	12.7 $\pm$ 1.13
Day 0 collar	1.3 $\pm$ 0.14	22.4 $\pm$ 0.99	3.3 $\pm$ 0.14	11.0 $\pm$ 0.71	47.0 $\pm$ 2.62	12.7 $\pm$ 2.69
Day 20 5°C back	1.2 ^b	23.7 $\pm$ 0.64	2.9 ^b	11.3 $\pm$ 0.28	50.1 $\pm$ 1.48	12.9 $\pm$ 2.40
Day 20 5°C collar	1.4 ^c	25.0 ^c	2.5 ^c	13.4 ^c	45.8 ^c	10.8 ^c
Day 20 15°C back	1.3 $\pm$ 0.07	23.4 $\pm$ 1.06	3.1 $\pm$ 0.35	11.6 $\pm$ 0.78	47.3 $\pm$ 0.21	12.6 $\pm$ 2.69
Day 20 15°C collar	1.4 $\pm$ 0.21	23.0 $\pm$ 1.84	3.4 $\pm$ 0.57	10.9 $\pm$ 1.06	46.6 $\pm$ 1.91	14.4 $\pm$ 0.14
<u>DRY SALT CURE</u>						
Day 0 back	1.4 $\pm$ 0.07	24.2 $\pm$ 1.27	3.3 $\pm$ 0.42	13.1 $\pm$ 0.49	45.7 $\pm$ 2.26	12.4 $\pm$ 0.64
Day 0 collar	1.3 $\pm$ 0.28	23.3 $\pm$ 0.99	3.3 $\pm$ 0.49	11.0 $\pm$ 0.07	46.8 $\pm$ 0.28	14.4 $\pm$ 0.57
Day 20 5°C back	0.6 $\pm$ 0.28	23.0 $\pm$ 0.21	3.1 $\pm$ 0.35	11.2 $\pm$ 0.14	49.2 $\pm$ 1.77	13.1 $\pm$ 1.06
Day 20 5°C collar	1.1 ^b	23.4 $\pm$ 1.70	2.8 $\pm$ 0.42	12.2 $\pm$ 1.70	47.4 $\pm$ 0.71	13.8 $\pm$ 1.63
Day 20 15°C back	1.2 $\pm$ 0.28	23.9 $\pm$ 1.34	2.9 $\pm$ 0.42	12.3 $\pm$ 0.71	47.0 $\pm$ 1.48	12.8 $\pm$ 0.42
Day 20 15°C collar	1.4 $\pm$ 0.07	23.1 $\pm$ 2.26	3.2 $\pm$ 0.99	11.6 $\pm$ 2.90	46.6 $\pm$ 3.54	12.9 $\pm$ 0.21

^aAverages of 2 samples representative of the treatment and sampling period.

^bArea of peak of 1 sample was < 1% of total area.

^cValue represented by 1 sample - second sample not available due to sample shortages.

Appendix 6. Average sensory scores of dry-cured Wiltshire bacon processed without α -tocopherol.

Ranking ^a					
Bacon Sample	Flavor	Color Intensity	Mouth Feel	Saltiness	Overall Acceptability
<u>back bacon</u>					
Day 0	1.7	0.1	-0.7	2.1	1.2
Day 5 5°C	0.6	0.4	-1.4	1.3	0.2
Day 5 15°C	0.9	-0.5	-0.4	1.5	0.7
Day 10 5°C	0.4	-0.3	-0.6	1.1	0.4
Day 10 15°C	0.8	-1.0	0.6	1.4	0.8
Day 15 5°C	0.7	-0.2	-1.3	1.2	0.4
Day 15 15°C	0.7	-0.9	-0.3	0.7	0.6
Day 20 5°C	1.0	-0.1	-1.2	1.0	0.6
Day 20 15°C	0.9	-0.7	-0.2	1.3	0.4
<u>collar bacon</u>					
Day 0	1.8	1.2	1.0	1.7	1.8
Day 5 5°C	1.4	-0.1	1.4	1.3	1.4
Day 5 15°C	0.9	0.8	0.8	0.9	0.9
Day 10 5°C	0.6	1.2	0.1	1.1	0.4
Day 10 15°C	0.7	0.8	0.6	1.6	0.6
Day 15 5°C	0.9	0.3	0.4	1.0	0.9
Day 15 15°C	1.2	0.3	1.2	1.3	1.4
Day 20 5°C	0.8	0.8	1.3	1.2	0.8
Day 20 15°C	0.3	0.6	1.2	1.1	0.5

^aScale: Color Intensity, Texture and Saltiness:

0 = ideal

3 = very dark, dry or salty

-3 = very light, moist or undersalty.

Flavor Acceptability, Overall Acceptability:

0 = neutral

3 = very desirable

-3 = very undesirable

Appendix 7. Average sensory scores of immersion-cured Wiltshire bacon processed without α -tocopherol.

Ranking ^a					
Bacon Sample	Flavor	Color Intensity	Mouth Feel	Saltiness	Overall Acceptability
<u>back bacon</u>					
Day 0	0.6	-1.1	-0.7	1.8	0.3
Day 5 5°C	0.8	-0.7	0.0	1.5	0.6
Day 5 15°C	1.2	-1.0	0.0	1.3	1.1
Day 10 5°C	0.9	-0.1	-0.8	1.0	0.6
Day 10 15°C	0.2	-0.4	-1.0	1.7	0.1
Day 15 5°C	0.4	0.0	-1.3	1.6	0.4
Day 15 15°C	0.2	-0.3	-1.2	1.4	-0.1
Day 20 5°C	0.6	-0.8	-0.7	1.0	0.4
Day 20 15°C	1.4	-0.4	0.0	1.2	1.1
<u>collar bacon</u>					
Day 0	1.4	0.5	1.1	1.1	1.2
Day 5 5°C	0.5	1.1	0.7	1.8	0.5
Day 5 15°C	0.9	1.6	-0.2	1.6	0.8
Day 10 5°C	1.2	0.8	0.7	1.0	1.3
Day 10 15°C	0.6	1.2	1.0	0.8	0.7
Day 15 5°C	0.3	1.6	-0.2	1.0	0.3
Day 15 15°C	0.5	1.1	0.5	1.1	0.7
Day 20 5°C	0.5	0.8	1.2	0.8	0.6
Day 20 15°C	0.7	0.9	1.4	0.5	0.7

^aScale: Color Intensity, Texture and Saltiness:
 0 = ideal
 3 = very dark, dry or salty
 -3 = very light, moist or undersalty.
 Flavor Acceptability, Overall Acceptability:
 0 = neutral
 3 = very desirable
 -3 = very undesirable

Appendix 8. Average sensory scores of dry-cured Wiltshire bacon containing α -tocopherol.

Ranking ^a					
Bacon Sample	Flavor	Color Intensity	Mouth Feel	Saltiness	Overall Acceptability
<u>back bacon</u>					
Day 0	1.4	-0.6	0.2	0.9	1.0
Day 5 5°C	1.3	-0.5	-0.8	0.8	0.8
Day 5 15°C	1.3	-0.6	-0.3	0.6	1.0
Day 15 5°C	0.7	-0.8	-0.3	1.1	0.4
Day 15 15°C	0.3	-0.6	-0.2	0.9	0.1
Day 20 5°C	0.7	-0.8	-0.6	0.7	0.6
Day 20 15°C	1.0	-0.5	-0.2	0.3	0.7
<u>collar bacon</u>					
Day 0	1.0	0.8	1.0	1.6	0.8
Day 5 5°C	0.5	1.2	-0.1	1.4	0.6
Day 5 15°C	0.7	0.7	0.7	0.9	0.7
Day 15 5°C	0.6	0.9	0.7	1.0	0.7
Day 15 15°C	1.0	1.3	1.2	0.7	1.1
Day 20 5°C	1.1	1.1	0.5	1.1	1.0
Day 20 15°C	0.7	1.0	0.8	0.9	0.7

^aScale: Color Intensity, Texture and Saltiness:
 0 = ideal
 3 = very dark, dry or salty
 -3 = very light, moist or undersalty.
 Flavor Acceptability, Overall Acceptability:
 3 = neutral
 3 = very desirable
 -3 = very undesirable

Appendix 9. Average sensory scores of immersion-cured Wiltshire bacon containing α -tocopherol.

Ranking ^a					
Bacon Sample	Flavor	Color Intensity	Mouth Feel	Saltiness	Overall Acceptability
<u>back bacon</u>					
Day 0	0.9	-0.3	0.0	1.4	0.8
Day 5 5°C	0.9	-0.8	-0.7	1.0	0.6
Day 5 15°C	0.4	-0.7	-0.5	0.9	0.3
Day 15 5°C	1.0	-0.6	-0.4	0.9	0.6
Day 15 15°C	1.4	-0.7	0.0	0.8	1.2
Day 20 5°C	1.1	1.1	1.0	0.9	1.1
Day 20 15°C	0.6	0.7	1.0	0.5	0.5
<u>collar bacon</u>					
Day 0	1.7	0.4	1.4	0.8	1.2
Day 5 5°C	1.1	0.8	0.4	0.5	1.1
Day 5 15°C	0.7	1.2	0.8	0.6	0.7
Day 15 5°C	1.1	1.1	1.0	0.4	1.1
Day 15 15°C	0.5	1.0	0.7	0.5	0.4
Day 20 5°C	0.6	-0.2	-1.1	0.9	0.3
Day 20 15°C	0.5	-0.8	-0.1	1.4	0.4

^aScale: Color Intensity, Texture and Saltiness:
 0 = ideal
 3 = very dark, dry or salty
 -3 = very light, moist or undersalty.
 Flavor Acceptability, Overall Acceptability:
 0 = neutral
 3 = very desirable
 -3 = very undesirable

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Appendix 10. Total viable counts of microorganisms during processing on dry-cured Wiltshire bacon cured without α -tocopherol.

Side Identification	Total Viable Count/g ($\log_{10}$)	Side Identification	Total Viable Count/g ($\log_{10}$)
<u>After Brine Injection</u>			
1 - rind	4.4	1 - pleura	4.1
2 - rind	3.9	2 - pleura	3.5
3 - rind	4.3	3 - pleura	3.9
4 - rind	4.4	4 - pleura	4.0
	$\bar{x} = 4.3$		$\bar{x} = 3.9$
<u>After Maturation Period</u>			
1 - rind	3.2	1 - pleura	3.0
2 - rind	2.9	2 - pleura	2.9
3 - rind	3.8	3 - pleura	3.0
4 - rind	4.0	4 - pleura	2.6
	$\bar{x} = 3.5$		$\bar{x} = 2.9$

^aAverages represent duplicate samples in serial dilutions.

Appendix 11. Total viable counts of microorganisms during processing on immersion-cured Wiltshire bacon cured without α -tocopherol.

Side Identification	Total Viable Count/g ($\log_{10}$)	Side Identification	Total Viable Count/g ($\log_{10}$)
<u>After Brine Injection</u>			
1 - rind	4.1	1 - pleura	3.8
2 - rind	4.3	2 - pleura	3.9
3 - rind	4.4	3 - pleura	3.9
4 - rind	4.3	4 - pleura	3.9
	$\bar{x} = 4.3$		$\bar{x} = 3.9$
<u>After Immersion Period</u>			
1 - rind	4.8	1 - pleura	4.5
2 - rind	4.4	2 - pleura	4.0
3 - rind	4.4	3 - pleura	4.0
4 - rind	4.1	4 - pleura	4.3
	$\bar{x} = 4.4$		$\bar{x} = 4.2$
<u>After Maturation Period</u>			
1 - rind	3.5	1 - pleura	4.3
2 - rind	4.2	2 - pleura	3.5
3 - rind	4.0	3 - pleura	3.1
4 - rind	4.1	4 - pleura	3.0
	$\bar{x} = 4.0$		$\bar{x} = 3.5$

^aAverages represent duplicate samples in serial dilutions.

Appendix 12. Total viable counts of microorganisms during processing on dry salt-cured Wiltshire bacon cured with α -tocopherol.

Side Identification	Total Viable Count/g ($\log_{10}$)	Side Identification	Total Viable Count/g ($\log_{10}$)
<u>After Brine Injection</u>			
5 - rind	3.8	5 - pleura	3.3
6 - rind	3.1	6 - pleura	3.0
7 - rind	3.2	7 - pleura	3.3
8 - rind	3.3	8 - pleura	3.3
	$\bar{x} = 3.4$		$\bar{x} = 3.2$
<u>After Maturation Period</u>			
5 - rind	2.9	5 - pleura	3.2
6 - rind	2.9	6 - pleura	3.3
7 - rind	2.5	7 - pleura	3.1
8 - rind	2.7	8 - pleura	3.1
	$\bar{x} = 2.8$		$\bar{x} = 3.2$

^aAverages represent duplicate samples in serial dilutions.

Appendix 13. Total viable counts of microorganisms during processing on immersion-cured Wiltshire bacon cured with α -tocopherol.

Side Identification	Total Viable Count/g ($\log_{10}$)	Side Identification	Total Viable Count/g ($\log_{10}$)
<u>After Brine Injection</u>			
5 - rind	4.0	5 - pleura	3.5
6 - rind	3.7	6 - pleura	3.9
7 - rind	3.5	7 - pleura	3.7
8 - rind	3.8	8 - pleura	3.9
	$\bar{x} = 3.8$		$\bar{x} = 3.8$
<u>After Immersion Period</u>			
5 - rind	3.2	5 - pleura	3.9
6 - rind	3.8	6 - pleura	3.9
7 - rind	3.5	7 - pleura	3.4
8 - rind	3.6	8 - pleura	3.8
	$\bar{x} = 3.5$		$\bar{x} = 3.8$
<u>After Maturation Period</u>			
5 - rind	3.3	5 - pleura	2.6
6 - rind	3.0	6 - pleura	2.7
7 - rind	2.8	7 - pleura	2.4
8 - rind	2.8	8 - pleura	2.5
	$\bar{x} = 3.0$		$\bar{x} = 2.6$

^aAverages represent duplicate samples in serial dilutions.

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