and assayed for rubber content and biomass (Table III). As shown, the total rubber content of the controls increased 24%, while sample V increased from 55 to almost 96 g/plant, which was a 74% increase within a period of 120 days.

There are two possible explanations why only sample V showed an increase in rubber content. Four successive monthly applications of the bioregulator might be necessary to induce rubber accumulation. However, this would seem unlikely from past results. It is also possible that only one application of DCPTA might be necessary but that the time of year of application is critical. This would seem to be the case here. Applying this information to the results in Table II, it appears that the extra applications of DCPTA were not responsible for the increase rubber induction in the 90- and 120-day samples.

Manipulation of the sample V data in Table III showed that the increased total rubber level was the result of a larger than average plant size. The average dry weight was 957 g/plant for the sample V treatment as compared with that of 624 g/plant for the average of all other samples. This represented a 53% increase in biomass. While it seems appropriate to conclude that DCPTA caused a stimulation of biomass accumulation, as reported previously (Yokoyama et al., 1984), there is the possibility that other factors such as nutrient and environmental conditions contributed to the increased plant size. Since the plants in this experiment were planted within a single block, it was not possible to fully evaluate the relative contributions of bioregulation and environmental factors to the biomass increase. However, the conclusions mentioned above regarding single spray applications would seem justified.

Cultivar 11634 responded more readily to rubber induction via DCPTA and its analogues than did other cultivars. Therefore, it would seem likely that there might be other guayule cultivars that are more responsive than those that have been studied. A study of those characteristics of cultivar 11634 that differentiate it from less responsive guayule cultivars could help to lead this selection.

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# Investigation of Nitrite and Nitrate Levels in Paper Materials Used To Package Fresh Meat

Fiona Scriven, Peter Sporns,\* and Fred Wolfe

A method was developed for the analysis of nitrite and nitrate in paper using reduction to nitric oxide and chemiluminescence detection. The potential for fresh meat contamination by nitrite and nitrate from over 50 different samples of paper and paperboard packaging materials was evaluated by this method of analysis. Concentrations of nitrite were determined to be less than 5  $\mu$ g of NO<sub>2</sub><sup>-</sup>/g, with two exceptions containing 14.5 and 19  $\mu$ g of NO<sub>2</sub><sup>-</sup>/g. Nitrate concentrations were generally higher, though 50% of the materials analyzed contained less than 10  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g. The greatest hazard of contamination of fresh meat was determined to be through contact with gummed paper tape, which contains extremely high levels of nitrate (25 800–32 700  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g). Migration studies using giblet bags containing 120  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g in contact with chicken breast meat showed that low but detectable migration of nitrate from the paper packaging material to meat could occur at even these low nitrate levels.

The problems associated with carcinogenic nitrosamines in the food supply has lead to a great deal of scientific investigation in the last two decades (Magee and Barnes, 1967; Archer, 1982; Crosby, 1983). Recently paper packaging materials have been found to contain the nitrosamine *N*-nitrosomorpholine, which could migrate to foods (Hotchkiss and Vecchio, 1983; Hoffman et al., 1982; Sen and Baddoo, 1986). It is probable that paper packaging materials that contain nitrosamines would also contain nitrite and/or nitrate. Also disconcerting is evidence that even highly purified papers such as filter paper can contain nitrite (Fiddler and Gentilcore, 1975) and nitrate (Fawcett et al., 1976).

Although paper packaging materials have been investigated for nitrosamines, there has been no study into levels of nitrite and nitrate that might be present in food contact paper packaging materials. Packaging materials contaminated with nitrite or nitrate would be a major concern when used as wrappings for fresh meat, since moisture from the meat could extract these ions. With the many natural nitrosatable amines and amides present in meat (Singer and Lijinsky, 1976; Spinelli et al., 1974; Velisek et

Food Science Department, University of Alberta, Edmonton, Alberta T6G 2P5, Canada.

al., 1975), plus the high temperatures used in cooking, there would be a risk of nitrosamine formation in the cooked meat.

As well as nitrosamine concerns, nitrite and nitrate in fresh meat could lead to a residual pink color (cured meat color) in cooked meat. This residual pink color has been a problem in poultry products and may be due to formation of nitrosylmyoglobin after contamination from nitrite or nitrate (Froning, 1983). In some cases it has been suspected that unscrupulous processers of fresh meat may also attempt to retain the red color of fresh meat by addition of nitrite or nitrate. For the above reasons most regulatory agencies demand that fresh meat be essentially free of nitrites and nitrates.

It has been shown that chemiluminescence detection of nitric oxide produced by chemical reduction of nitrite and nitrate has advantages over spectrophotometric methods (Walters et al., 1979, 1980; Cox, 1980; Doerr et al., 1981; Fiddler et al., 1984) in many systems. These advantages include ease of analysis, sensitivity, reproducibility, and freedom from interference from suspended material and color. We set out, then, to develop a chemiluminescence method of analysis for nitrite and nitrate levels in paper materials and to use this method to examine the extent of nitrite and nitrate contamination in paper packaging materials used for fresh meat products.

# MATERIALS AND METHODS

All reagents used were reagent grade or better.

Paper packaging materials examined were obtained from local (Edmonton, Alberta, Canada) retail stores, distributers, and manufacturers. A large variety of paper packaging materials was also supplied by Agriculture Canada.

Method of Extraction of Paper. Paper packing samples (5.0 g) were cut into small pieces and blended to a pulp with 60 mL of water in a Waring blender. Whenever there was sufficient paper (10 g or more) two samples were carried through this procedure. The pulp was transferred quantitatively to a 250-mL flask, and water was added to make a total solution of 150 mL. After refluxing for 30 min, the paper solution was cooled to room temperature and filtered through a sintered-glass filter. The total volume of filtrate, with washings of the reflux flask and paper, was made up to 200 mL. Twenty-milliliter aliquots of the solution were analyzed, in duplicate, for both nitrite and total nitrite and nitrate. If the nitrite or nitrate content of the sample was above the upper range limit for analysis, a smaller aliquot was diluted to 20 mL and analyzed.

Method of Extraction of Chicken Meat. Skinned chicken breast (10.0 g) was cut into small pieces and homogenized with 60 mL of water with a Waring blender. Two samples were prepared from each chicken breast. The meat slurry was transferred to a 250-mL flask, and water was added to make a total volume of 150 mL. The solution was heated on a steam bath for 1 h, cooled to room temperature, and filtered through a sintered-glass filter. The total volume of filtrate, with washings of flask and meat sample, was made up to 200 mL. Twenty-milliliter aliquots of the solution were analyzed, in duplicate, for both nitrite and total nitrite and nitrate.

Analytical System. A 20-mL aqueous sample was placed in a 100-mL three-necked round-bottomed flask. One neck of the flask was connected to a nitrogen flow maintained at a pressure of 1 atm, another was fitted with an injection port containing a Teflon-lined rubber septum, and the third neck was connected to a Bendix 8101-C Oxides of Nitrogen (NO<sub>x</sub>) analyzer via a 5 M sodium hydroxide trap and a trap containing 5 g of anhydrous calcium sulfate (nonindicating). The two traps were used to remove acidic fumes and water vapor.

Nitrite Analysis. Nitrogen gas was passed through the solution for 5 min before the sample was treated with 1 mL of 0.2 M sodium iodide and 3 mL of glacial acetic acid. Nitric oxide produced from reduction of nitrite was drawn from the reaction vessel in a stream of nitrogen. Nitric oxide evolution was monitored by the NO<sub>x</sub> analyzer interfaced with an integrator (Hewlett-Packard Model 3390A). This was continued until no further nitric oxide evolution was observed, the area under the nitric oxide peak in combination with the relevant range factor of the NO<sub>x</sub> analyzer being proportional to the concentration of nitrite in the sample.

A linear nitrite calibration curve was prepared for a series of 20-mL sodium nitrite standard solutions over the concentration range of  $0.005-10.000 \ \mu g$  of  $NO_2^-$  (r = 0.9993) so that quantitative evaluation of samples could be obtained by comparison of peak areas. The limit of detection (2:1 signal-to-noise ratio) was  $0.005 \ \mu g$  of  $NO_2^-$  in a 20-mL sample.

Nitrate Analysis. After nitrogen was passed through the sample for 5 min, 1 mL of 5% w/v titanium(III) chloride solution and 5 mL of concentrated sulfuric acid were added. The nitric oxide produced was detected as described above for nitrite.

A linear nitrate calibration curve was prepared for a series of 20-mL nitrate standard solutions over the concentration range of  $0.12-40.00 \ \mu g$  of  $NO_3^-$  (r = 0.9926) so that again quantitative evaluation of samples could be obtained by comparison of peak areas. The limit of detection (2:1 signal-to-noise ratio) was  $0.12 \ \mu g$  of  $NO_3^-$  in a 20-mL sample.

Under the nitrate reduction conditions, nitrite and nitrate are reduced in equimolar amounts. Nitrate concentration of a sample can be calculated as the difference between total nitrite and nitrate content, and the nitrite content, determined as described previously.

Migration Experiments Using Chicken Breasts. Unpackaged processed chickens were obtained directly from a local poultry processor (Lilydale, Edmonton). The breast meat was removed from the carcass and skinned. Half of the breast was brought into one-sided contact with an 11 by 12 cm section of giblet bag (1.5 g) contained in a 15-cm diameter petri dish. The sample was covered and stored for 5 days at refrigeration temperatures. The other half of the breast was placed in another petri dish and stored in the same way. Five chicken breasts were treated in this manner. A further five samples and controls were frozen for 1 week and then thawed at 4 °C for 5 days. All samples were analyzed for nitrite and nitrate after the storage period.

### **RESULTS AND DISCUSSION**

We originally investigated a sequential method of analysis for nitrite and nitrate introduced by Walters et al. (1979; 1980), since this method determined nitrite and nitrate directly (without extraction) from food or other matrices. Using the apparatus described by Walters et al. (1980), we were unable to obtain a nitric oxide response on our NO<sub>x</sub> analyzer for amounts of nitrite lower than 20  $\mu$ g in 0.5 mL of water or methanol. Also, reproducibilities and limits of detection were not satisfactory for analysis of low levels of nitrite so an alternate method was sought.

We settled on a modification of the method of analysis originally used by Cox (1980) and later by other workers (Garside, 1982; Yoshizumi et al., 1985). Reproducibility of our method was found to be comparable (Table I) with similar methods of nitrite and nitrate analysis reported by

 Table I. Concentration of Standard Solutions Determined

 by the Chemiluminescence Method of Analysis

nitrite concn, µg NO2 <sup>-</sup> /L	nitrite in 20-mL sample anal., μg	nitrite concn determined, μg NO <sub>2</sub> <sup>-</sup> /L	coeff of variation, %
8.0	0.160	8.6 ± 0.7	8
23.9	0.478	$23.7 \pm 0.2$	1
31.8	0.636	$31.5 \pm 1.6$	5
100.0	2.000	$101.0 \pm 2.5$	2
nitrate	nitrate in 20-mL	nitrate concn	
concn, μg NO <sub>3</sub> <sup>-</sup> /L	sample anal., µg	determined, μg NO <sub>3</sub> <sup>-</sup> /L	coeff of variation, %
,		,	
$\mu g NO_3^{-}/L$	anal., µg	μg NO <sub>3</sub> -/L	variation, %
μg NO <sub>3</sub> <sup>-</sup> /L 25.0	anal., μg 0.50	$\frac{\mu \text{g NO}_3^-/\text{L}}{26.7 \pm 1.7}$	variation, % 6

 Table II. Recovery of Sodium Nitrite and Potassium

 Nitrate Added to Paper Packaging Materials

nitrite added, µg	nitrite rec,ª µg	mean rec, μg	% rec	
100.0	112.0 109.5	$100.3 \pm 12.3$	$100 \pm 12$	
	$\begin{array}{c} 87.4\\92.3\end{array}$			
30.0	$29.2 \\ 28.8$	27.2 • 2.6	91 ± 9	
	$\begin{array}{c} 27.3\\ 23.6\end{array}$			
nitrate	nitrate	mean		
added, µg	$rec,^a \mu g$	rec, µg	% rec	
100.0	114.0 97.5	$102.7 \pm 9.4$	$103 \pm 9$	
	106.2 92.9			
30.0	29.1 37.5	$28.9 \pm 6.1$	<b>96 ±</b> 20	
	24.9			

<sup>a</sup> Means of duplicate analyses.

others (Cox, 1980; Doerr et al., 1982; Fiddler et al., 1984). Peak half-widths were of the order of 2.5 min, and the total analysis time per sample was 20 min for either nitrite or nitrate. A shorter peak width could be obtained using a smaller (smaller than 20-mL) sample, but because of dilution of paper samples on extraction, it was preferable to analyze a large sample to obtain sufficient reproducibility and sensitivity.

Recovery of nitrite and nitrate was studied on a freezer wrap and a peach paper, determined as containing low levels of nitrite and nitrate by an alternate analysis procedure (Association of Official Analytical Chemists, 1984) as well as with the chemiluminescence method of detection. It should be noted that the former procedure could not be used for colored paper because extracted color interfered in this colorimetric analysis. The recoveries of added nitrite and nitrate at levels equivalent to 6 and 20 ppm nitrite and nitrate were investigated with these paper materials (Table II). The values obtained for these duplicate analyses were comparable to values obtained by Fiddler et al. (1984) for meat samples. The errors in the recovery values result from initial errors in the determination of nitrite and nitrate background levels and from variations between different samples of the same packaging material.

Fifty-six different samples of paper and paperboard packaging materials were analyzed for nitrite and nitrate by the developed chemiluminescence method of detection.

Table III. Nitrite and Nitrate Levels in Paper Materials Used in Packaging Fresh Meat

	$\operatorname{concn}^{b} \mu g/g$	
type of paper <sup>a</sup>	nitrite	nitrate
butcher's kraft (7)	1.0, 0.1-4.1	25.1, 3.7-60.6
locker wrap (5)	0.3, 0.1 - 0.7	5.7, 2.5-7.7
white oiled paper (3)	0.5, 0.4-0.7	10.8, 5.2-14.5
vegetable parchment (3)	1.0, 0.6-2.1	20.5, 5.3-46.8
peach paper (6)	2.0, 0.7 - 4.4	24.7, 4.5-77.1
meat and poultry pads (5)	4.6, 0.3-14.5	9.6, 1.9-18.9
gummed tape (4)	1.1, 0.4 - 2.5	28 100, 25 800-32 700
freezer tape (1)	1.1	6.2
cellulose tape (1)	0.2	1.5
paper tape (1)	0.7	6.6
masking tape (2)	1.1, 0.8 - 1.3	3.0, 2.9-3.1
cellotape (1)	0.8	1.5
giblet bags (3)	1.8, 1.1-2.4	47.8, 3.8-119.8
brown packing paper (1)	1.6	19.5
corregated cardboard (2)	2.5, 1.3 - 3.7	40.7, 42.5-38.8
cardboard and plastic trays (5)	5.1, 1.4-19	26.5, 14.0-47.9
cardboard and foil tray (1)	2.8	2.1
cardboard holder (1)	0.9	162.4
cardboard and polyethylene lid for freezer-pak (1)	3.5	37.0
cardboard freezer-pak (3)	1.9	14.3
polystyrene tray (1)	0.2	3.4
polyethylene giblet bag (1)	0.3	10.3
stamp (1)	2.0	7.1
envelope flap (1)	2.1	10.4

<sup>a</sup> Number of samples analyzed in parentheses. <sup>b</sup> Average levels in samples analyzed followed by range of values.

The overall mean concentrations of nitrite and nitrate detected in the packaging materials and the range of values observed are shown in Table III.

With the exception of two samples, levels of nitrite in paper packaging products were less than 5 ppm. One sample of poultry pads was anomalous with a concentration of nitrite of 14.5 ppm, as was one cardboard and plastic tray with a concentration of nitrite of 19.0 ppm.

Concentrations of nitrate were more variable from sample to sample and ranged from 1.5 to 32700 ppm. The highest concentrations of nitrate were found in gummed paper tape at 25800-32700 ppm (25.8-32.7 mg of NO<sub>3</sub><sup>-</sup>/g). These extremely high concentrations are due to the use of sodium nitrate in the manufacture of starch-based adhesives (Jarowenko, 1977; Kirby, 1965; Shields, 1984).

The base paper for gummed tape is usually a strong kraft grade specially sized to prevent undue penetration of the adhesives. The water-remoistenable adhesive is a dried film of adhesive material originally laid down from water and activated by moistening with water. Animal and vegetable glues and dextrins are most commonly used with the addition of modifiers that impart specific properties such as flexibility, good spreading, ease of rewetting, etc. Sodium nitrate is added to starch at levels of about 5-20% to act as a plasticizer, humectant, and/or liquefier (Jarowenko, 1977). Plasticizers provide the adhesive bond with flexibility, liquefiers reduce the viscosity of the adhesive, and humectants control the drying rate and absorb moisture. Sodium nitrate is used as a low-cost additive to adhesives, but there are alternatives that can be used. Urea imparts the same properties as sodium nitrate to the adhesive (Jarowenko, 1977; Shields, 1984). Other humectants and liquefiers include calcium chloride, thiourea, acetamide, and guanidine salts. Other plasticizers include glycerol, glycol, sorbitol, glucose, sugar, and corn syrup.

Nevertheless, nitrate was found at high concentrations in all samples of gummed tape obtained from four different sources. The tape is commonly used to secure the outer wrap of a meat package. If the meat is wrapped poorly or if there is excessive drip loss from the meat, the outer tape could be wetted and since nitrate is a highly soluble ion, migration from the tape to the meat will almost certainly occur. This project was originally motivated by just such an incident, when a fresh steak which retained a reddish pink color on cooking was found to contain levels of nitrate of about 25 ppm (using Association of Official Analytical Chemists, 1984, procedure) due to leaching of nitrate from gummed tape.

Starch-based adhesives that may contain sodium nitrate are used in the manufacture of other packaging materials which have food applications such as corrugated board, paper bags, cartons, and paper boxes. Starch-based adhesives are also used for postage stamps, envelopes, and labels. For interest we analyzed some postage stamps and the adhesive flap of some envelopes, but concentrations of nitrate were less than 15  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g.

Other meat paper packaging products that may contain nitrate-modified adhesives were investigated during the course of this study. Meat and poultry pads consist of a polyethylene backing adhered to several layers of absorbent tissue.

Several samples were analyzed; concentrations were in the range of 1.9–18.9  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g and not of the same order as the gummed tape. Paper giblet bags contain some adhesive to form the bag. For three types of giblet bags, concentrations ranged from 3.8 to 119.8  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g. The giblet bags with a nitrate concentration of 119.8  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g contained a large amount of adhesive.

Nitrate concentrations were found to be relatively high in paper samples that do not contain adhesives. Some samples of peach paper, white, brown, and red kraft, vegetable parchment, and locker wrap contained nitrate at concentrations of  $30.4-60.6 \ \mu g$  of NO<sub>3</sub><sup>-</sup>/g. Higher nitrate levels, between 17.0 and 162.4  $\mu g$  of NO<sub>3</sub><sup>-</sup>/g, were detected in cardboard trays and corrugated cardboards that generally contain some adhesive. Twenty-eight samples of paper packaging materials contained less than 10  $\mu g$  of NO<sub>3</sub><sup>-</sup>/g.

Other adhesive tapes used in the meat industry such as freezer tape, cellulose tape, and masking tape were analyzed. A rubber resin adhesive system is used in these pressure-sensitive tapes. There is no evidence in the literature of nitrate being added to this type of adhesive, and concentrations were found to range from 1.5 to 6.6  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g and 0.2 to 1.3  $\mu$ g of NO<sub>2</sub><sup>-</sup>/g accordingly.

With the exception of the gummed tape, the giblet bags containing 119.8  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g were considered to present the highest probability of contamination to fresh meat. Giblet bags can remain within a bird for up to 5 days at refrigeration temperatures and for several months in the frozen product. On thawing, or in fresh birds, the body cavity in which the giblet bag is contained is extremely aqueous with blood and drip loss from the bird. Since nitrate is a highly water-soluble ion, the body cavity of the bird presents an ideal environment for migration of nitrate from the giblet bags could also be a contributory factor to the residual pink problem sometimes encountered in processed poultry products.

To investigate the above possibility, an experiment was performed on chicken breast muscle. First, recoveries of nitrite and nitrate from chicken breast muscle were determined with  $100 \ \mu g$  of  $NO_2^-$  or  $100 \ \mu g$  of  $NO_3^-$  added to a meat slurry prior to extraction. The extraction solutions were analyzed for nitrite and nitrate by the chemiluminescence method of detection. The nitrite and nitrate contents of the unadulterated chicken sample were subtracted to obtain the recovery values. For a chicken breast sample with 100  $\mu$ g of NO<sub>2</sub><sup>-</sup>, added as sodium nitrite, the mean recovery for four analyses was 89.9 ± 2.1  $\mu$ g of NO<sub>2</sub><sup>-</sup>, or about 90% recovery. For a chicken breast sample with 100  $\mu$ g of NO<sub>3</sub><sup>-</sup>, added as potassium nitrate, the mean recovery for four analyses was 97.3 ± 10.2  $\mu$ g of NO<sub>3</sub><sup>-</sup>, or about 97% recovery. For these samples mean recoveries again compared favorably with those obtained by Fiddler et al. (1984) for a meat sample fortified with 5 ppm nitrite.

Evidence of migration of nitrite or nitrate from the giblet bag to the meat was obtained for only 2 of the 10 samples analyzed. One sample showed a low migration of 0.04  $\mu g$ of  $NO_2^{-}/g$  of chicken breast and a higher 0.97  $\mu g$  of  $NO_3^{-}/g$ of chicken, while another sample had a level of nitrate migration of 0.78  $\mu$ g of NO<sub>3</sub>/g of chicken. Nitrate contamination was much higher (about 25 times) than nitrite contamination since nitrate concentrations in the giblet bags were about 120  $\mu$ g of NO<sub>3</sub><sup>-</sup>/g and nitrite concentrations were about 2  $\mu g$  of NO<sub>2</sub><sup>-</sup>/g. It is unlikely that nitrite and nitrate contamination at these levels from packaging materials would be a major cause of residual pink color in processed poultry products, though it may still be a minor contributory factor. Preslaughter stress, nitrate in chill water, and exhaust fumes from transportation trucks are considered to be the major factors (Froning, 1983). However, migration from paper to fresh meat of, especially nitrate, has been demonstrated. With paper packaging materials containing higher levels of nitrites and nitrates contamination could be a concern. For example, similar migration of nitrate from gummed paper tape, with about 300 times the levels of nitrate, would result in levels of up to 200 ppm nitrate.

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**Registry No.**  $NO_3^-$ , 14797-55-8;  $NO_2^-$ , 14797-65-0; polystyrene, 9003-53-6; polyethylene, 9002-88-4.

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# Effect of Cesium-137 Radiation on the Formation of N-Nitrosopyrrolidine in Bacon

John W. Pensabene,\* Robert A. Gates, Ronald K. Jenkins, and Walter Fiddler

To date, the majority of information collected on the irradiation of bacon has been based on  ${}^{60}$ Co as the radiation source. In this study,  ${}^{137}$ Cs was used as the radiation source for the treatment of bacon. Bacon prepared with 550 ppm sodium ascorbate and either 120 or 40 ppm sodium nitrite was irradiated at absorbed doses of 0.75, 1.5, and 3.0 Mrad. Results show that only at 3.0 Mrad was there a significant difference from the nonirradiated control in both residual nitrite and *N*-nitrosopyrrolidine (NPYR) values and that NPYR levels were significantly less in the 40 ppm nitrite bacon than in the 120 ppm bacon. Radiation of bacon prepared with and without ascorbate also showed an additive effect on nitrite destruction and NPYR reduction. The results suggest  ${}^{137}$ Cs may not be as effective as  ${}^{60}$ Co in reducing nitrite/NPYR in bacon.

Nitrite, in combination with sodium chloride, is used in the curing of meat to produce its characteristic color and flavor and to control spoilage and growth of pathogenic microorganisms. In particular, the control of *Clostridium* botulinum has been claimed to be very important with respect to bacon to justify the use of 120 ppm ingoing sodium nitrite  $(NaNO_2)$ . The problem with this is that the higher the ingoing nitrite, the higher the residual nitrite content. For bacon, there is a high correlation between residual nitrite prior to frying and N-nitrosopyrrolidine (NPYR), a known animal carcinogen, after frying. In addition to nitrosamines, the question concerning whether nitrite itself is carcinogenic or not was raised as a result of the Newberne study (Newberne, 1979). While the interpretation of this study's results has now been largely discounted, two recent papers have revised the question of the toxicological significance of nitrite itself. First, the study by Lijinsky et al. (1983), in which nitrite was fed to rats for 2 years, indicated that nitrite increased the incidence of liver neoplasms in female, but not in male rats, and second, the report by Schweinsberg and Bürkle (1985) indicated that nitrite may be a cocarcinogen in the presence of other carcinogens. As a result, different approaches need to be considered to reduce the amount of nitrite used in processing cured meat products.

Radiation sterilization has been proposed as a possible means of reducing levels of nitrite in bacon to the minimum needed for the development of the characteristic cure color and flavor, and also for the requisite protection against C. botulinum. Anellis and Werkowski (1968) showed that C. botulinum spores are destroyed by irradiation at a dose between 2.0 and 2.9 Mrad. The only known drawback to the use of radiation in bacon is that the process has not yet been approved for use by the Food and Drug Administration (FDA).  $\gamma$  radiation has recently been approved for use to control insect infestation in certain species (Federal Register, 1985a) and to kill *Trichinella spiralis* in pork (Federal Register, 1985b).

In our earlier work on the effect of radiation sterilization (Fiddler et al., 1981), bacon prepared with 120 ppm NaNO<sub>2</sub> was irradiated with <sup>60</sup>Co (3.0 Mrad) at -40 °C. We found that residual NaNO<sub>2</sub> was reduced to almost nondetectable levels prior to frying and that NPYR after frying was less than the USDA violative level of 10 ppb. We also found that, in bacon prepared with either 20 or 40 ppm NaNO<sub>2</sub>, radiation yielded NPYR values indistinguishable from nitrite-free bacon. In the present work, we carried out a series of experiments on NANO<sub>2</sub>/NPYR in bacon to determine whether <sup>137</sup>Cs would give us the same trends we obtained earlier with <sup>60</sup>Co (Fiddler et al., 1981).

# EXPERIMENTAL SECTION

**Reagents.** A complete list of reagents needed for determining NPYR in fried bacon and its cooked-out fat was reported elsewhere (Pensabene and Fiddler, 1982; White et al., 1974).

Bacon Processing. Skinned, matched pork bellies were purchased from a local supplier within 24 h postmortem and stored at -18 °C until used. Prior to use, the bellies were thawed for 1 week in a cooler at 1 °C. All bellies were pumped to approximately 110% of green weight to achieve ingoing levels of 1.5% sodium chloride, 0.75% sucrose, 0.3% sodium tripolyphosphate, 0 or 550 ppm sodium ascorbate (NaAsc), and 40 or 120 ppm NaNO<sub>2</sub>. The pumped bellies were stored in polyethylene bags at 1 °C for 18 h and then processed in a smokehouse as described previously (Pensabene et al., 1979). After processing, the bellies were chilled overnight in a 1 °C cooler and then sliced. The randomized slices were vacuum packaged (12 slices/pkg) prior to irradiation at 0.75, 1.5, and 3.0 Mrad with a  $^{137}Cs$ source and a chamber temperature of 2-3 °C. A complete description of the irradiator, including source strength,

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U.S. Department of Agriculture, Philadelphia, Pennsylvania 19118.