

Thermal Decomposition of the Rubber Vulcanization Agent, Zinc Dibenzylthiocarbamate, and Its Potential Role in Nitrosamine Formation in Hams Processed in Elastic Nettings

John S. Helmick and Walter Fiddler*

Eastern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, Philadelphia, Pennsylvania 19118

The thermal decomposition of zinc dibenzylthiocarbamate (ZnDBzDTC), a compound used in the formulation of rubber and a possible precursor for *N*-nitrosodibenzylamine (NDBzA), was studied by a variety of thermal and spectroscopic techniques. At 326 °C, the decomposition temperature of the dithiocarbamate, carbon disulfide and dibenzylamine were the principal products formed. Smaller amounts of toluene, benzyl isothiocyanate, *N,N,N'*-tribenzylthiourea, and benzylbenzylidene were identified. The amount of dibenzylamine (DBzA) formed by the thermal decomposition of ZnDBzDTC may have a limited role in the formation of NDBzA in hams processed in elastic rubber nettings. The thermal conditions used in the smokehouse are significantly lower than the decomposition temperature of purified ZnDBzDTC.

Keywords: *Thermal decomposition; dithiocarbamate; rubber; dibenzylamine; nitrosamines; cured meats*

INTRODUCTION

The first report of nitrosamines associated with rubber and rubber products was the discovery of both *N*-nitrosodimethylamine and *N*-nitrosomorpholine in the air of rubber and tire manufacturing plants (Fajen et al., 1979). These same two nitrosamines were also detected in the air from the interior of new automobiles, with the maximum amount found in the spare tire well area (Fine et al., 1980). Another study (McGlothlin et al., 1981) involving a single tire manufacturing plant found these same two nitrosamines plus *N*-nitrosopyrrolidine and *N*-nitrosodiphenylamine in the factory air. About the same time, there were several disclosures of nitrosamines in rubber products, other than those associated with automobile tires. These included rubber stoppers in blood collection tubes (Lakritz and Kimoto, 1980), rubber gloves, condoms, and baby bottle nipples (Ireland et al., 1980), and rubber toys, gloves, balloons, and water hoses (Spiegelhalder and Preussmann, 1982). The finding of nitrosamines in rubber nipples and pacifiers was a major concern because of potential infant exposure to these carcinogenic compounds. These products were studied extensively, resulting in regulatory action (Novitch, 1983; Hile, 1984) that forced the manufacturers to reformulate the rubber so as not to produce nitrosamines.

While nitrosamines in rubber nipples were shown to migrate into milk and formula during heat sterilization conditions (Havery and Fazio, 1982), Sen et al. (1987) reported the first example of a food product containing nitrosamines by direct contact with a rubber-containing material. *N*-Nitrosodibutylamine and a smaller amount of *N*-nitrosodiethylamine were found in Canadian cured meat products processed in elastic rubber netting. In a later paper (Sen et al., 1988), these same researchers found *N*-nitrosodibenzylamine (NDBzA) rather than *N*-nitrosodibutylamine in hams, indicating that the netting manufacturers were using reformulated rubber.

This was recently confirmed by Pensabene and Fiddler (1992). The carcinogenicity of *N*-nitrosodibutylamine prompted the use of an accelerator that formed an apparent noncarcinogenic nitrosamine, NDBzA (Druckrey et al., 1967), although there is evidence of this nitrosamine's genotoxicity (Schmezer et al., 1987; Boyes et al., 1990). All of the literature on this subject, from the earliest findings to the present, attributes the formation of the nitrosamines to the accelerators used in rubber production (Ireland et al., 1980; Yeager et al., 1980). The compounds used for this purpose are dithiocarbamates, thiuram mono- and polysulfides, sulfenamides, and thioureas. They are typically made from secondary amines and sold as unpurified bulk chemicals. As a result, rubber can contain nitrosatable amines both as a contaminant of and as a decomposition product from the accelerator (R. F. Ohm, R. T. Vanderbilt Co., personal communication, 1993). Zinc dibenzylthiocarbamate (ZnDBzDTC), rather than the dibutyl derivative, is currently the principal accelerator being used in the formulation of the rubber used for the elastic netting.

Nitrosamines are present in the highest concentrations on the exterior surface of the hams (Sen et al., 1987). Under the thermal processing conditions normally used in the smokehouse, the temperature is higher at the ham surface than in the interior. This, and the intimate contact between the nitrite-containing pork and the elastic rubber netting, have prompted the study of the mechanism of formation of rubber-related nitrosamines in pork. In this paper we report the results obtained from the thermal decomposition of purified ZnDBzDTC to determine the extent of amine precursor formation.

MATERIALS AND METHODS

Safety Note. Caution should be exercised in the handling of nitrosamines, since they are potential carcinogens.

The dibenzylthiocarbamic acid sodium salt and carbon disulfide (CS₂) were supplied by Aldrich Chemical Co., Milwaukee, WI. The zinc sulfate heptahydrate (ZnSO₄·7H₂O) was

* Author to whom correspondence should be addressed.

supplied by Mallinckrodt Inc., Paris, KY. The dibenzylamine (DBzA) was supplied by Janssen Chimica, Spectrum Chemical Manufacturing Corp., New Brunswick, NJ. All water used in this study was first deionized and then distilled in an all glass apparatus, while solvents were of high purity HPLC UV grade supplied by Burdick & Jackson, Muskegon, MI.

Apparatus and Procedures. The synthesis of ZnDBzDTC was carried out using a modification of the procedure of Martens and Githens (1952). Dibenzylthiocarbamic acid sodium salt (5.91 g) was dissolved in 200 mL of water. In a separate flask, 2.88 g of zinc sulfate heptahydrate was dissolved in 200 mL of water with stirring. When the zinc sulfate was dissolved, the solution of carbamic acid was slowly added, with continual stirring, over a period of 1 h. As the reaction progressed, a white precipitate formed, which was filtered off 60 min after the addition of the carbamic acid. The filtrate was washed with cold distilled deionized water and then with cold acetone. The sample was recrystallized from acetone. The identity and purity of ZnDBzDTC were confirmed by high-resolution NMR and elemental analysis.

The differential scanning calorimetry (DSC) curves for ZnDBzDTC were recorded using a Perkin-Elmer Model DSC-7 differential scanning calorimeter at a heating rate of 10 °C/min using sample weights ranging from 2 to 4 mg. The temperature range in which the sample was analyzed was between 30 and 420 °C.

The thermal gravimetric analysis (TGA) of ZnDBzDTC was carried out using a DuPont Model 950 thermal gravimetric analyzer with computer interface and software designed by Micro-Scan Services, Inc., Schenectady, NY. The samples, weighing between 10 and 12 mg, were heated under a nitrogen atmosphere from 70 to 500 °C with a temperature program of 5 °C/min.

The thermal gravimetric analysis-Fourier transform infrared (TGA-FTIR) experiments were carried out using the same instrument and software as indicated above. The sample, weighing 10–13 mg, was heated from 210 to 460 °C at 5 °C/min in an atmosphere of flowing nitrogen. The IR analysis was carried out using an Analect RFX-30 FTIR equipped with a high-sensitivity mercury-cadmium-thallium (MCT) detector. The effluents from the TGA were directed through transfer lines heated to 250 °C into a 2 mm × 10 cm light pipe. The spectra were collected at 15 s intervals over a time period of 50 min with 200 scans averaged at 4 cm⁻¹ resolution.

The gas chromatograph-mass spectrometer (GC-MS) injector temperature study was carried out by following a method similar to that reported by Muller and Stan (1990) for determination of the thermal stability of several carbamate pesticides. A series of triplicate 1 µL injections of a known amount of ZnDBzDTC [1 mM in dichloromethane (DCM)] were made into the GC, over a range of injector temperatures from 200 to 300 °C. The subsequent increase in peak area of DBzA with increasing temperature was noted. A series of concentrations of DBzA, in DCM, was also run in triplicate for the calibration curve. This was used to calculate the percent DBzA formed at each injector temperature from the ZnDBzDTC decomposition response. The study was carried out on a Hewlett-Packard Model 5890A gas chromatograph equipped with a Model 5971 mass selective detector (MSD). Gas chromatograph conditions: splitless injector temperatures started at 200 °C and then were increased in 10 °C increments to 300 °C; injector purge gas off for 5 min; helium (grade 5.0) carrier gas, 1.2 mL min⁻¹. Column and conditions: 30 m × 0.25 mm i.d., 0.25 µm, DB-17 capillary column; program, 70 °C for 5 min, 70–250 °C at 9 °C/min, then 250 °C for 20 min. Mass spectrometer conditions: detector temperature, 300 °C; GC-MS interface, 200 °C; purge on at 2 min and off at 5 min; MS vacuum, 50–70 mTorr; scan rate, 0.6 scans/s; mass scan range, 15–600 amu.

RESULTS AND DISCUSSION

The DSC curve for ZnDBzDTC, obtained over a temperature range of 30–420 °C, showed two thermal events. Figure 1 shows an expanded version over the

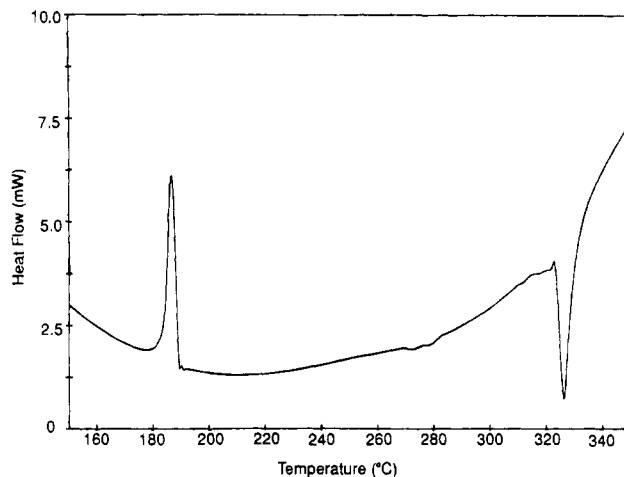


Figure 1. Differential scanning calorimetric analysis of ZnDBzDTC.

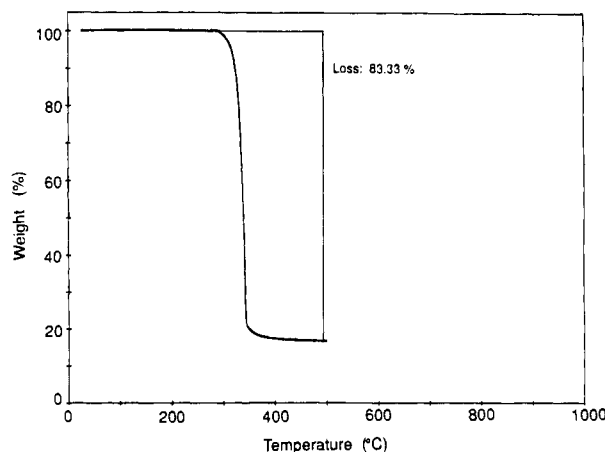


Figure 2. Thermal gravimetric analysis of ZnDBzDTC.

range from 150 to 350 °C where they occur. The first event at 186 °C was endothermic and is characteristic of the phase change at the melting point of the ZnDBzDTC. The second event at 326 °C was exothermic. The latter fell within the 300–330 °C temperature range of the TGA curve shown in Figure 2. The TGA curve shows only one significant weight loss, at a level of about 83%.

In an effort to characterize the decomposition products as they were liberated, a TGA was run between 210 and 460 °C and the IR spectrum collected every 15 s. A waterfall plot of the spectral data over the 295–330 °C range is shown in Figure 3. The predominant peaks at ca. 1500 and 2200 cm⁻¹ are characteristic of carbon disulfide (CS₂). This was confirmed by obtaining a spectrum of an authentic sample of CS₂. Carbon disulfide also had the same elution as the compound obtained from the decomposition of ZnDBzDTC. Subtraction of the spectra for CS₂ showed that other unidentified organic components were being evolved with CS₂ at the same temperature (Figure 4). The peaks at 3100 cm⁻¹ are aromatic C–H stretch bands, and the broad peaks between 2850 and 2950 cm⁻¹ are aliphatic C–H stretches. The broad peak at 2100 cm⁻¹ suggests that a isocyanate, thiocyanate, or isothiocyanate may be present. Medium-sized peaks centered at 1200 and 1530 cm⁻¹ are indicative of a thiocarbonyl group linked to a nitrogen, thereby denoting that one of the compounds is an isothiocyanate. This class of compounds typically shows C–N triple-bond or double-bond stretches in the 2270–2000 cm⁻¹ region (Silver-

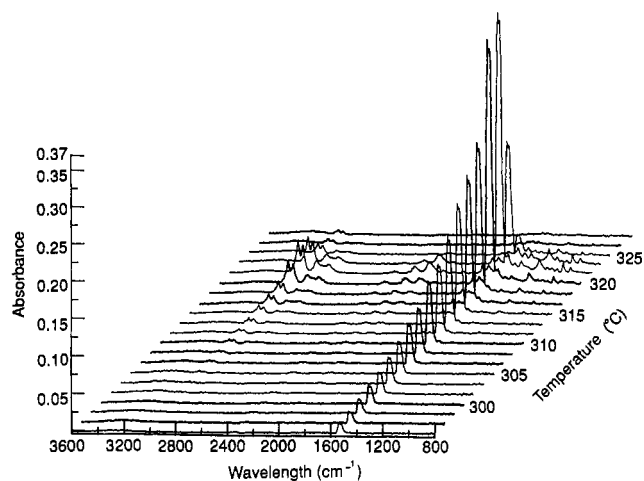


Figure 3. Waterfall plot of the full IR spectrum of the TGA evolve decomposition products over the temperature range 295–330 °C.

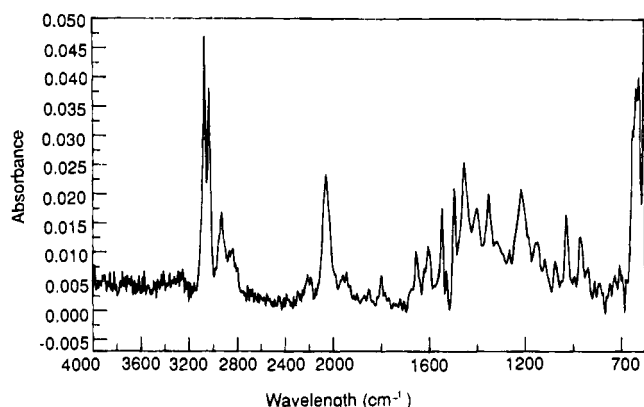


Figure 4. Full IR spectrum taken at 320 °C after subtraction of the carbon disulfide spectrum.

stein et al., 1981). The overall spectrum indicated that a mixture of organic components was obtained, as would be expected from the thermolysis of carbamate derivatives (Fernandez-Alba et al., 1992). There was no evidence of DBzA, thought to be one of the other primary decomposition products. This conclusion was reached when none of the major peaks in the spectra mixture matched those obtained for DBzA. The temperature of the interface between the TGA and FTIR could not be heated to a temperature high enough to volatilize DBzA (bp 268–271 °C at 250 mmHg) so it could pass into the FTIR light pipe. Alternately, the DBzA could be decomposing in the heated metal interface. For this reason, a separate study was carried out on the formation of DBzA from ZnDBzDTC. In it, the amount of DBzA was monitored by GC-MS as the injection port temperature was varied. On-column injection of ZnDBzDTC minimized exposure to a metal surface. Figure 5 shows the percent of DBzA formed from ZnDBzDTC as a function of the injector temperature. At 200 °C only trace quantities (<1%) of DBzA were formed. As the injector temperature was increased, the amount of DBzA also increased. Temperatures in excess of 300 °C could not be employed for this study due to concern about the possible harmful effects to the mass spectrometer's injector and column. Comparison of the DBzA formation curve with the TGA ZnDBzDTC curve indicated that the temperature at which the greatest rate of decomposition occurs would also correspond to where most of the DBzA forms. This was the 326 °C decomposition temperature obtained in

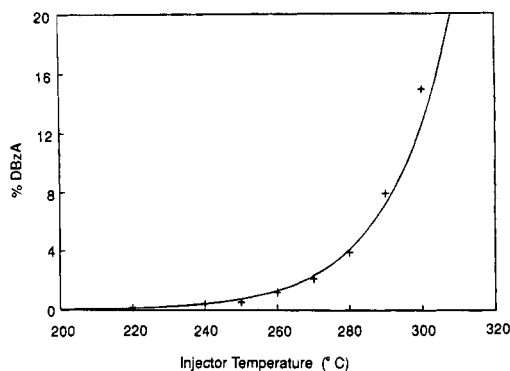


Figure 5. Curve of dibenzylamine formation (percent) versus GC injector temperature; detection by MSD.

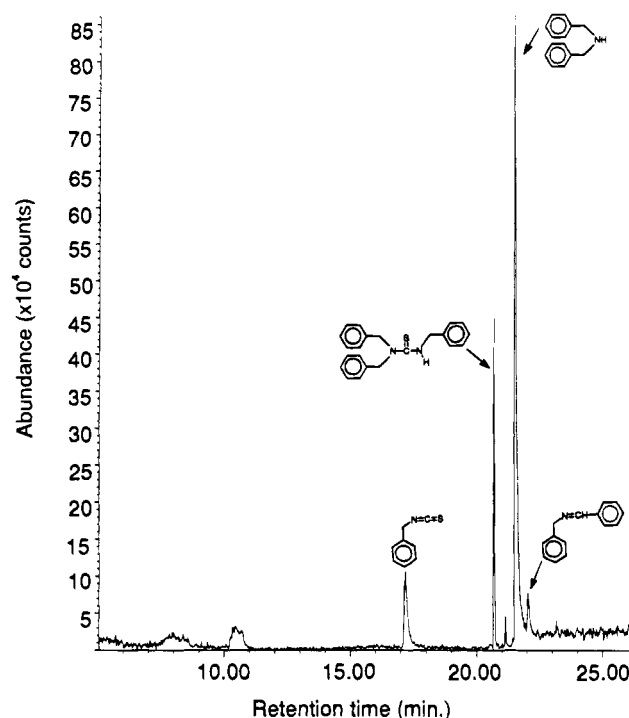


Figure 6. GC-MS total ion chromatogram of ZnDBzDTC decomposition at 300 °C.

both the DSC and TGA studies. These results indicate that at this temperature both carbon disulfide and DBzA are evolved. Total ion spectra were taken at the same injector port temperature increments at the same time DBzA was monitored. For example, at 270 °C, two small peaks were detected. In Figure 6 taken at 300 °C, these two peaks at 17.2 and 20.8 min were significantly higher at this temperature, as was the DBzA at 22.7 min. The two compounds were identified as benzylisothiocyanate and *N,N,N'*-tribenzylthiourea, respectively. The small peak after DBzA at 22.1 min was identified as benzylidene benzylamine. This shows that the thermal decomposition of ZnDBzDTC is not a simple one where only CS₂ and DBzA are formed. These two decomposition products could be readily formed by thermolysis at the C-N bond [N-C(=S)] and at the S-Zn bond. Homolysis at the benzyl C-N bond could produce toluene, which we detected under other chromatographic conditions. Cleavage at the S-C bond in S-C(=S) explains the formation of benzyl isothiocyanate. It, in turn, could react with DBzA to form the tribenzylthiourea. Another secondary reaction product, benzylidene benzylamine, was most likely formed from benzaldehyde and benzylamine. Pyrolysis at 400 °C and elemental analysis of the residue indicated that zinc

sulfide (ZnS) was the major nonvolatile constituent. The 82% weight loss shown by TGA (ca. 18% residue, nonvolatile material) is within 2% of the expected residual of 16%, if all of the nonvolatile material was ZnS. Elemental analysis indicated a minor amount of carbon-containing material, similar to that noted by Cui et al. (1992) for the decomposition of nickel, cobalt, and zinc diethyldithiocarbamate complexes.

The DBzA is thought to be formed during the rubber vulcanization process, in which temperatures can reach 160–180 °C and, in some processes, as high as 200 °C. This study shows that these temperatures are significantly lower than the temperature required for optimum ZnDBzDTC decomposition–DBzA formation. Dibenzylamine may also be formed upon extended heating time at a temperature lower than the maximum decomposition temperature. This is related to the formation of NDBzA in boneless hams processed in elastic nettings where ZnDBzDTC is used in the formulation of the rubber. In commercial practice, the smokehouse temperature does not exceed 220 °F (104 °C), but the nitrated cured pork can be exposed to the elastic rubber netting for up to 12 h. Therefore, a study of the thermal decomposition of ZnDBzDTC was carried out at this temperature for up to a 24 h, with the sample flask swept with nitrogen into a bubble tube containing 2 N HCl. No significant weight loss of ZnDBzDTC or detection of DBzA was observed.

In conclusion, the results suggest that the amount of DBzA available for nitrosation by thermal decomposition may be limited and has a minor role in nitrosamine formation. However, other sources of this nitrosamine precursor may be available, for example, DBzA formed by the hydrolysis of ZnDBzDTC and the unreacted amine that may be present after the accelerator's synthesis. Five different samples of rubber were separated from unused elastic nettings extracted with DCM and the extracts concentrated and analyzed using an injector temperature of 160 °C. Dibenzylimine was detected in all samples; the results ranged from 34.2 to 200.3 ppm. The thermal decomposition characteristics of purified ZnDBzDTC suggest there are substantial amounts of undecomposed accelerator still present in the rubber after the vulcanization process. This is with the recognition that a number of compounds used in the formulation of rubber facilitate the decomposition of ZnDBzDTC. Therefore, the content of DBzA as a contaminant in the industrial grade vulcanizing agent and residual accelerator in the rubber warrants further investigation.

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LITERATURE CITED

- Boyes, B. G.; Rogers, C. G.; Matula, T. I.; Stapley, R.; Sen, N. P. Evaluation of genotoxicity of N-nitrosodibenzylamine in Chinese hamster V79 cells and in Salmonella. *Mut. Res.* **1990**, *241*, 379–385.
- Cui, H.; Pike, R. D.; Kershaw, R.; Dwight, K.; Wold, A. Synthesis of nickel sulfide, cobalt sulfide, and zinc sulfide by the decomposition of diethyldithiocarbamate complexes. *J. Solid State Chem.* **1992**, *101*, 115–118.
- Druckrey, H.; Preussmann, R.; Ivankovic, S.; Schmahl, D. Organotropic carcinogenic activity of 65 different N-nitroso compounds on BD-rats. *Z. Krebsforsch.* **1967**, *69*, 103–201.
- Fajen, J. M.; Carson, G. A.; Rounbehler, D. P.; Fan, T. Y.; Vita, R.; Goff, U. E.; Wolf, M. H.; Edwards, G. S.; Fine, D. H.; Reinhold, V.; Bieman, K. N-Nitrosamines in the rubber and tire industry. *Science* **1979**, *205*, 1262–1264.
- Fernandez-Alba, A.; Perez-Alvarez, I. J.; Martinez-Vidal, J. L.; Gonzalez-Pradas, E. Thermal and spectroscopic study of several dithiocarbamates. *Thermochim. Acta* **1992**, *211*, 271–277.
- Fine, D. H.; Reisch, J.; Rounbehler, D. P. N-Nitrosamines in new automobiles. In *N-Nitroso Compounds: Analysis, Formation and Occurrence*; Scientific Publication 31; International Agency for Research on Cancer (IARC): Lyon, France, 1980; pp 541–551.
- Havery, D. C.; Fazio, T. Estimation of volatile N-nitrosamines in rubber nipples for babies' bottles. *Food Cosmet. Toxicol.* **1982**, *20*, 939–944.
- Hile, J. P. *Fed. Regist.* **1984**, *49*, 50789–50790.
- Ireland, C. B.; Hytrek, F. P.; Lasoski, B. A. Aqueous extraction of N-nitrosamines from elastomers. *Am. Ind. Hyg. Assoc.* **1980**, *41*, 895–900.
- Lakritz, L.; Kimoto, W. N-Nitrosamines: contaminants in blood collection tubes. *Food Cosmet. Toxicol.* **1980**, *18*, 31–34.
- Martens, R. I.; Githens, R. E. Small amounts of copper in dyes and rubber chemicals: colorimetric determination with zinc dibenzylthio-carbamate. *Anal. Chem.* **1952**, *24*, 991–993.
- McGlothlin, J. D.; Wilcox, T. C.; Fajan, J. M.; Edwards, G. S. A health hazard evaluation of nitrosamines in a tire manufacturing plant. In *Chemical Hazards in the Workplace*; ACS Symposium Series 149; American Chemical Society: Washington, DC, 1981; pp 283–299.
- Muller, H.-M.; Stan, H.-J. Thermal degradation observed with different injection techniques: quantitative estimation by the use of thermolabile carbamate pesticides. *J. High Resolut. Chromatogr.* **1990**, *13*, 759–763.
- Novitch, M. *Fed. Regist.* **1983**, *48*, 57014.
- Pensabene, J. W.; Fiddler, W.; Gates, R. A. Solid phase extraction method for volatile nitrosamines in hams processed in elastic rubber nettings. *J. Assoc. Off. Anal. Chem.* **1992**, *75*, 438–442.
- Schmezer, P.; Pool, B. L.; Preussmann, R.; Schmahl, D. Analysis of N-nitrosamines for genotoxicity in primary hepatocytes derived from various species. In *The Relevance of N-Nitroso Compounds to Human Cancer: Exposure and Mechanisms*; Scientific Publication 84; IARC: Lyon, France 1987; pp 270–273.
- Sen, N. P.; Baddoo, P. H.; Seaman, S. W. Volatile nitrosamines in cured meats packaged in elastic rubber nettings. *J. Agric. Food. Chem.* **1987**, *35*, 346–350.
- Sen, N. P.; Seaman, S. W.; Baddoo, P. H.; Weber, D. Further studies on the formation of nitrosamines in cured pork products packaged in elastic rubber nettings. *J. Food Sci.* **1988**, *53*, 731–734.
- Silverstein, R. M.; Bassler, G. C.; Morrill, T. C. *Spectrometric Identification of Organic Compounds*, 4th ed.; Wiley: New York, 1981; pp 166–170.
- Spiegelhalder, B.; Preussmann, R. Nitrosamines in rubber. In *N-Nitroso Compounds: Occurrence and Biological Effects*; Scientific Publication 41; IARC: Lyon, France, 1982; pp 231–243.
- Yeager, F. W.; van Gulick, N. N.; Lasoski, B. A. Dialkyl nitrosamines in elastomers. *Am. Ind. Hyg. Assoc. J.* **1980**, *41*, 148–150.

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