NITRITES AND NITROSAMINES IN PROCESSED MEATS

EFFECT OF INITIAL LEVEL OF CONTAMINATION AND PROCESSING TEMPERATURES ON FINAL BOLOGNA PRODUCT CHARACTERISTICS AND MICROBIAL LOAD

H. W. OCKERMAN, R. F. PLIMPTON, Jr., and D. W. LONG

The Ohio State University, Columbus, Ohio 43210 and The Ohio Agricultural Research and Development Center, Wooster, Ohio 44691, U.S.A.

This research investigated in a comminuted cooked product the interrelationships between initial microbial load (5.3 vs. 7.0/g), processing internal temperatures (62.8°C, 68.3°C and 73.9°C) and processing holding times after achieving internal temperatures (0 and 15 minutes). To follow microbial changes, microbiological examinations were conducted on the meat blocks, the uncooked emulsions and the finished cooked products. Other evaluations included yield, reflectance and a sensory panel rating for color, texture, juiciness, bologna flavor, off-flavor and general acceptability. Initial contamination level was determined to be the most important factor in producing a low microbial bologna product with desirable quality attributes. An increase in internal cooking temperature reduced the microbial numbers but also lowered the general quality of the cooked product. A 15-minute holding time at the temperatures used was not sufficient to significantly alter the microbiological level of the finished product.

EFFET DU NIVEAU INITIAL DE CONTAMINATION ET DES TEMPERATURES DE TRAITEMENT SUR LES CARACTERISTIQUES FINALES DE LA SAUCISSE DITE "BOLOGNA" ET SA CHARGE MICROBIENNE.

H. W. OCKERMAN, R. F. PLIMPTON, JR. , et D. W. LONG.

Ohio State University, Columbus, Ohio 43210 en collaboration avec le centre pour la recherche et le développement de l'agriculture, Wooster, Ohio 44691, U.S.A.

Ce travail de recherche a eu pour but l'étude dans un produit réduit par la cuisson des relations entre la charge microbienne initiale (5,3g. contre 7,0g.), les températures intérieures de traitement (62°8C, 68°3C et 73°9C) et les durées de traitement après avoir atteint les températures internes de traitement (0 à 15 mn). Pour suivre les développements microbiens, des expériences microbiologiques ont été effectuées sur des bouts de viande, des émulsions crues et des produits préalablement cuits. D'autres études ont apprécié la teneur, la réflectibilité et une estimation sensorielle de la couleur, la texture, le caractère juteux, la saveur particulière à la saucisse dite "bologna", le goût désagréable de ce produit-ci et l'acceptabilité globale. Le niveau initial de contamination fut considéré le facteur le plus important pour la production d'une saucisse à bas niveau microbien et possédant les caractéristiques désirables. Une augmentation de la température intérieure de cuisson a réduit les éléments microbiens mais a aussi diminué la qualité générale du produit cuit. 15 minutes de cuisson aux températures utilisées ne furent pas necessaires pour alterer d'une manière significative le niveau microbiologique du produit final.

AUSWIRKUNGEN DER ANFANGSVERSEUCHUNG UND VERARBEITUNGSTEMPERATUREN AUF DAS ENDPRODUKT BOLOGNA-WURST UND DIE MIKROBISCHE BELASTUNG

H. W. OCKERMAN, R. F. PLIMPTON, JR., und D. W. LONG

The Ohio State University, Columbus, Ohio 43210 und The Ohio Agricultural Research and Development Center, Wooster, Ohio 44691, USA.

Diese Forschung untersuchte die Verhältnisse in einem zerriebenen gekochten Erzeugnis zwischen der Anfangsmikrobialbelastung (5,3 vs. 7,0/g), der inneren Verarbeitungstemperaturen (62,8°C, 68,3°C, und 73,9°C), und der weiteren Verarbeitungszeiten nach Erreichung der inneren Temperaturen (0 bzw. 15 Minuten). Um mikrobische Veränderungen festzustellen, wurden die Fleischwürfel, die ungekochten Emulsionen und das fertige Produkt mikrobiologisch untersucht. Andere Untersuchungen gaben Endvolumen, Reflexionsstärke, sowie Resultate der Sinneneindrücke von Farbe, Beschaffenheit, Saftigkeit, Wurstgeschmack, verdorbenem Geschmack und allgemeine Annehmbarkeit wieder. Der Anfangsverseuchungsgrad wurde als wichtigster Faktor in der Erzeugung eines Produktes mit niedrigem mikrobischen Grad und den gewünschten Qualitätsattributen festgestellt. Die Zunahme der inneren Verarbeitungstemperaturen hat die mikrobische Zahl reduziert aber auch die allgemeine Qualität des gekochten Erzeugnisses vermindert. Eine 15-minutige Verarbeitung war bei den angewandten Temperaturen nicht ausreichend, um den mikrobiologischen Anteil des Endproduktes bedeutend zu verändern.

ВЛИЯНИЕ ИСХОДНОГО УРОВНЯ СОДЕРЖАНИЯ МИКРОБОВ И РАЗЛИЧНЫХ ТЕМПЕРАТУР ПРИ ОБРАБОТКЕ НА СВОЙСТВА ГОТОВОГО ПРОДУКТА БОЛОНСКОЙ КОЛБАСЫ И СОДЕРЖАНИЕ МИКРОБОВ В НЕЙ

Х. В. Оккерман, Р. Ф. Плимптон, младший, и Д. В.Лонг, Государственный университет штата Огайо, Колумбус, Огайо, 43210 и Агрономический научно-исследовательский центр штата Огайо, Вустер, Огайо, 44691, США.

В этом исследовании была изучена зависимость в размельченном варенном продукте между исходным содержанием микробов (5.3 и 7.0 в одном грамме), внутренней температурой обработки (62.8 Ц, 68.3 Ц и 73.9 Ц) и продолжительностью обработки после достижения внутренней температуры (0 и 15 минут). Чтобы наблюдать изменения в содержании микробов, микробиологические исследования производились в кусочках мяса, в неваренной эмульсии и в готовом варенном продукте. Были исследованы также изменения в весе, отражение света и комиссия из нескольких человек произвела непосредственную оценку цвета, свойства ткани, сочности, вкуса болонской колбасы, нежелательного постороннего привкуса и общей приемлемости. Было установлено, что для изготовления болонской колбасы с желательными свойствами и низким содержанием микробов самым важным фактором является исходный уровень содержания микробов. Увеличение внутренней температуры варения понижало количество микробов, но это также понижало качество варенного продукта. Продолжительность времени в 15 минут при использованных температурах не была достаточной, чтобы существенно изменить уровень содержания микробов в готовом продукте.

NITRITES AND NITROSAMINES IN PROCESSED MEATS

ARTICLE FOR XX EUROPEAN MEAT RESEARCH CONFERENCE (1974)

EFFECT OF INITIAL LEVEL OF CONTAMINATION AND PROCESSING TEMPERATURES ON FINAL BOLOGNA PRODUCT CHARACTERISTICS AND MICROBIAL LOAD

H. W. Ockerman, R. F. Plimpton, Jr. and D. W. Long, The Ohio State University, Columbus, Ohio 43210 and The Ohio Agricultural Research and Development Center, Wooster, Ohio 44691 U.S.A.

INTRODUCTION

Recent consumer pressures (Consumer Reports, 1972) have focused attention on the need for renewed efforts by the meat industry to reduce microbial levels in comminuted products. The industry is aware that controls on the level of bacterial contamination are being suggested and that continuous economic loss due to microbiological spoilage can not be tolerated. Two basic approaches seem possible. One is to alter the number of microorganisms in the starting material and the second is to reduce this number by processing techniques.

PROCEDURE

This experiment was designed to investigate the interrelationship between initial microbial level, processing internal temperatures, and processing holding times after achieving internal temperatures. The experimental design is illustrated in Table 1.

Table 1. EXPERIMENTAL DESIGN

Variables	Treatments
Initial Microbiological Loads	Normal (10^5 to 10^7/g): High (Normal plus and inoculated, more than 10^7/g)
Final Internal Processing Temperature	62.8°C, 68.3°C, 73.9°C
Holding Time at Final Internal Processing Temperature	0 minutes, 15 minutes

Twenty-four batches of bologna were produced with 12 containing the normal microbiological load. Tissue was obtained in a manner similar to that used commercially and the microbiological loads on this tissue were typical of those normally encountered in industry. The other 12 batches, in addition to the normal level, were inoculated with additional microorganisms (Labeled - high level). Of the 12 batches made from each initial contamination level, groups of four were cooked to one of three final internal temperatures (62.8°C, 68.3°C, or 73.9°C). Two of each group of 4 were removed from cooking immediately when the detected internal temperature was reached whereas 2 were held at this temperature for an additional 15 minutes.

Each meat block, emulsion, and individual stick of cooked product was analyzed for total bacterial count using three different incubation temperatures (22°C, 37°C, and 50°C). In addition each resulting stick of bologna was analyzed by percent reflectance (color), panel evaluation (color, texture, juiciness, bologna flavor, off-flavor, and general acceptability) and for percent yield.

All pork tissue was taken 80 hours post-mortem from the ham and shoulder area of one carcass for a given run and ground through a 6 mm breaking plate. One-half of this tissue was used for the preparation of the normal microbiological level meat block.

To produce the high level of contamination, one-half of the meat block was inoculated with 250 ml of 0.85% saline solution containing approximately 100,000 organisms per ml. The normal contamination sample was also mixed with 250 ml of sterile saline solution. The inoculum was prepared by using Pseudomonas putrefaciens (OSU #89), maintained on Bacto Nutrient Agar (Difco #B1), grown in Nutrient Broth (Difco #BC), centrifuged, and resuspended in 0.85% saline solution.

Batches of bologna were produced using the formulation shown in Table 2.

Table 2. BOLOGNA FORMULATION

Ingredient	Quantity (grams)
Boneless Pork (25% fat)	2,268.0
Ice	453.6
NaCl (salt)	70.0
Dextrose	23.0
White Pepper	7.5
Ground Coriander	3.5
Sage	1.5
Garlic Powder	1.0
Mace	1.0
Sodium Nitrate	0.5
Sodium Nitrite	0.4
Sodium Erythrobate	1.7
CHAR SOL (liquid smoke)	4.0 (ml)

The emulsion mixture was chopped approximately 2 minutes in a vertical chopper with the temperature not exceeding 18°C. The emulsion was then stuffed into presoaked 2 inch (Union Carbide fibrous, "Easy Peel") bologna casings to produce 2 two-pound sticks per batch.

The products were placed on smokehouse trees and cooked at an internal smokehouse temperature of 57°C for 30 minutes. The house temperature was raised to 85°C and retained at that setting until the internal temperature was 10 degrees below the treatment temperature. At that time the smokehouse temperature was lowered to the desired temperature and held there for the remainder of the cooking cycle. The product was removed when it reached the treatment temperature or held at that temperature for an additional 15 minutes according to the experimental design. In both cases the product was subjected to a 30-minute cold water shower prior to being placed in a 3°C cooler for a 24-hour storage period.

Microbial examinations of the meat block, uncooked emulsion, and finished cooked product were conducted using the standard pour plate technique for the enumeration of microorganisms. Plates were prepared using Tryptone Glucose Extract Agar (Difco) and duplicates at each dilution were incubated in each of 3 incubation temperatures. The 50°C and 37°C plates were incubated for 48 hours and the 22°C plates were incubated for 96 hours.

The percent yield was calculated by dividing the 24 hours out of the smokehouse weight by the uncooked emulsion weight.

Reflectance was determined on a Bausch and Lomb spectronic 20 equipped with a reflectance unit. The procedure used was similar to the technique described by Warnecke (1962) and consisted of a reflectance ratio taken at 650 mμ and 570 mμ.

A trained panel was used for the subjective evaluation of the finished product using a 1 to 10 hedonic scale with 10 being desirable and 1 being unacceptable.

RESULTS AND DISCUSSION

The effect of the initial level of contamination and the microbiological incubation temperatures on initial microbial count, fresh emulsion microbial count and final bologna product microbial count are shown in Table 3.

Table 3. THE EFFECT OF INITIAL MICROBIAL CONTAMINATION ON THE LEAST SQUARES MEANS FOR INITIAL, EMULSION, AND BOLOGNA MICROBIAL COUNTS AT THREE INCUBATION TEMPERATURES[1/ 2/]

	Normal Contamination	Normal Contamination plus Inoculum	Standard Error	Significance[3/]
INITIAL COUNT				
Incubated at 22°C	5.27	7.00	0.44	**
Incubated at 37°C	4.33	6.30	0.21	**
Incubated at 50°C	2.00	2.49	0.24	*
FRESH EMULSION COUNT				
Incubated at 22°C	5.27	5.66	0.27	N.S.
Incubated at 37°C	4.48	5.23	0.14	**
Incubated at 50°C	2.56	3.03	0.30	N.S.
BOLOGNA COUNT[4/]				
Incubated at 22°C	2.70	3.08	0.14	*
Incubated at 37°C	2.51	2.72	0.05	**
Incubated at 50°C	2.30	2.69	0.10	**

1/ Least square means

2/ Microbial count expressed as the log of the number of organisms per gram

3/ Significance: N.S.-non significant (P>.05), *-significant (P<.05), **-significant (P<.01)

4/ Least square means with cooking temperature and holding time removed

The microbial counts for the normal contamination level meat block were similar to those reported in the literature. Weiser et al. (1971) reported ground pork for sausage manufacturing to have greater than 1,000,000 organisms per gram while Heiszler et al. (1972) reported between 100,000 and 10,000,000 in the meat block for emulsion products. Warnecke (1962) reported the counts for his pork meat block to be between 10,000 and 10,000,000 per gram at a 30°C incubation. The counts decreased as incubation temperatures increased which can be explained by noting that the predominant organisms found in refrigerated meat are psychrophilic.

The inoculation was effective in increasing the number of microorganisms found at all 3 incubation temperatures. Due to the psychrophilic nature of the inoculum organisms, the magnitude of the increase at the 50°C incubation temperature was less than at lower incubation temperatures.

When comparing the microbial counts from the fresh emulsion product with that of the initial meat block counts, the results indicate there was a reduction in counts at the lower incubation temperatures and an increase at the higher incubation temperature. The addition of salt and nitrite would probably explain the lower incubation temperature results, particularly on the Pseudomonas putrefaciens inoculated samples. Preliminary work showed that the spice mixture contained more than 10,000 organisms per gram at the 50°C incubation temperature and their growth would account for the increase obtained at the 50°C incubation temperature. In the fresh emulsion product the inoculated samples contained more microorganisms than the comparable normal samples. Although this difference was not as great as in the initial meat block the difference in all cases did approach significance.

After the bologna was cooked the microbiological counts were reduced and this reduction was of increased magnitude as the incubation temperature was lowered. When comparing normal vs. inoculated samples under all incubation temperatures, the inoculated samples had a significant increase in bacterial numbers indicating that initial load was one of the factors determining final bacterial count even in a cooked product. These cooked microbial counts were slightly lower than the ones reported by Warnecke (1962, 1966) and Heiszler et al. (1974).

The data used to evaluate the microbiological growth relationships at different incubation temperatures, are shown in Table 4.

Table 4. MICROBIOLOGICAL COUNTS WHEN EVALUATION PLATES ARE INCUBATED AT DIFFERENT TEMPERATURES

	Correlation[1/] Coefficient
RAW MEAT PRODUCTS (n=36)	
Log # incubated at 22°C= 1.71 + 0.87 (log # incubated at 37°C)	0.69**
= 5.23 + 0.37 (log # incubated at 50°C)	0.25
Log # incubated at 37°C= 1.68 + 0.54 (log # incubated at 22°C)	0.69**
= 3.98 + 0.45 (log # incubated at 50°C)	0.38*
Log # incubated at 50°C= 1.11 + 0.16 (log # incubated at 22°C)	0.25
= 0.52 + 0.32 (log # incubated at 37°C)	0.38*
SALTED UNCOOKED MEAT EMULSION PRODUCTS (n=36)	
Log # incubated at 22°C= 2.16 + 0.70 (log # incubated at 37°C)	0.57**
= 3.64 + 0.61 (log # incubated at 50°C)	0.70**
Log # incubated at 37°C= 2.14 + 0.46 (log # incubated at 22°C)	0.57**
= 3.86 + 0.26 (log # incubated at 50°C)	0.37*
Log # incubated at 50°C=-1.52 + 0.81 (log # incubated at 22°C)	0.70**
= 0.43 + 0.53 (log # incubated at 37°C)	0.37*
SALTED COOKED MEAT EMULSION PRODUCTS (n=72)	
Log # incubated at 22°C= 1.25 + 0.61 (log # incubated at 37°C)	0.31**
= 2.11 + 0.28 (log # incubated at 50°C)	0.21*
Log # incubated at 37°C= 2.08 + 0.15 (log # incubated at 22°C)	0.31**
= 2.04 + 0.20 (log # incubated at 50°C)	0.30**
Log # incubated at 50°C= 1.93 + 0.16 (log # incubated at 22°C)	0.21*
= 1.26 + 0.44 (log # incubated at 37°C)	0.30**

1/ Significance: * - P<.05, ** - P<.01

NITRITES AND NITROSAMINES IN PROCESSED MEATS

Comparison of microbiological results when the evaluation plates are incubated at different temperatures can be made for raw meat products, uncooked emulsions and cooked emulsions using these prediction equations if the inherent problems of this type of comparisons are also considered.

Most research has suggested that initial contamination of the raw material has a significant influence on the quality of the cooked emulsion product even though a large percentage of these initial organisms are destroyed in the cooking process. Microbiologically initiated quality changes occurring prior to cooking, and products of microbial metabolism and heat resistant microorganisms have been suggested as the possible cause of this deterioration in quality.

Table 5 presents the effects of microbial contamination on cooked bologna appearance, yield and palatability.

Table 5. EFFECT OF INITIAL LEVEL OF CONTAMINATION ON THE LEAST SQUARES MEANS FOR APPEARANCE, YIELD, AND PANEL PALATABILITY EVALUATION[1/]

Evaluation	Normal Contamination	Normal Contamination plus Inoculum	Standard Error	Significance[2/]
Reflectance Ratio 650 mμ/570 mμ	1.87	1.89	0.06	N.S.
Yield, %	91.02	91.17	0.31	N.S.
Panel Color Score[3/]	6.87	7.02	0.17	N.S.
Panel Texture Score[3/]	7.39	7.14	0.11	*
Panel Juiciness Score[3/]	7.51	7.24	0.14	A.S.
Panel Bologna Flavor Score[3/]	7.45	7.28	0.10	A.S.
Panel Off-flavor Score[3/]	8.78	8.68	0.11	N.S.
Panel General Acceptability[3/]	7.27	7.05	0.13	A.S.

1/ Least square means

2/ Significance: N.S.-non-significant ($P > .1$), A.S. - approaches significance ($P < .1$), * - significance ($P < .05$)

3/ Panel scores on a 1 to 10 hedonic scale (10 - desirable; 1 - unacceptable)

Initial contamination level had no significant influence on yield or color as measured by reflectance or panel. The color data agrees with Warnecke et al. (1966). They found that microbial contamination prior to processing did not affect color development. The increased contamination of the raw material did produce a reduced rating for the inoculated samples when the panel compared these for texture, juiciness, bologna flavor, off-flavor, and general acceptability. All of these differences with the exception of off-flavor approached significance ($P < .10$). This again agrees with the research of Warnecke et al. (1966) who reported that initial microbial growth in the raw material had a very detrimental effect on texture, flavor and over-all desirability of the end product.

As the least squares means for the product with final internal temperatures of 63°, 68°, and 74°C were compared, the microbiological counts (22°C incubation) significantly ($P < .05$) decreased as the temperature increased. This reduction was from log 3.07/g to log 2.74/g when 63°C internal temperature was compared to 68°C and from 3.07/g to 2.53/g when 63°C was compared to 74°C. The reduction in bacterial numbers agrees with that found by Heiszler et al. (1972) who reported slight reductions in bacterial count at 30°C incubation for 10°F increase in final cooking temperature. The microbial counts were not significantly different when the plates were incubated at the higher temperatures. This would suggest that final internal cooking temperature can be used to reduce the microbial load of psychrophilic organisms in a comminuted product.

Panel color (7.41, 6.30, 6.26), juiciness (8.12, 7.05, 6.69) and off-flavor scores (9.17, 8.59, 8.50) all decreased significantly ($P < .05, < .01, < .05$, respectively) as initial temperature (63°, 68°, 74°C) was increased. General acceptability decreased (7.78, 6.83, 7.07) significantly ($P < .01$) from internal temperature of 63°C to 68°C but did not decrease from 68° to 74°C. Texture (7.46, 7.37, 7.15) and bologna flavor (7.85, 7.02, 7.01) scores approached a significant ($P < .10$) reduction as temperature was increased. The other comparisons were non-significant when compared on an internal temperature basis. This would suggest that most of the major indicators of product quality decrease as the internal temperature is increased from 63 to 74°C. Protein denaturation with increasing temperature would probably account for most of these reductions in panel scores as the temperature was increased. General acceptability yielded a highly significant correlation with panel color scores (0.58) and panel juiciness scores (0.69) helping to explain the high general acceptability value at the 63°C internal temperature point.

When the least squares means were compared for zero and 15 minutes holding time at each temperature or combined over-all temperatures the only significant ($P < .05$) difference was a reduction in yield (91.41%, 90.87%) with the longer holding time. Fifteen minutes was chosen as the holding time because it was suspected that this would be sufficient time to kill most psychrophilic organisms and would be sufficiently short to be compatible with industry's continuous processing procedures. The results of this experiment would indicate that 15 minutes at the temperatures used was not long enough to have a significant effect on bacterial reduction. This is also in agreement with Heiszler et al. (1972) who reported that holding the final internal temperature for up to 30 minutes had no sufficient effect on final microbial count.

CONCLUSIONS

Initial contamination level was the most important factor evaluated in producing a low microbial bologna product with desirable quality attributes. An increase in internal cooking temperature will reduce the microbial numbers but will also lower the general quality of the cooked product. A 15-minute holding time at the temperatures used was not sufficient to significantly alter the microbiological level of the finished product.

REFERENCES

Consumers Union of the United States, Inc. 1972. "Frankfurters". Consumer Reports 37:73.

Difco Manual. 1953. Difco Laboratories, Inc., Detroit, Michigan.

Heiszler, M.G., A. A. Kroft, C. R. Rey and R. E. Rust. 1972. Effect of Time and Temperature of Smoking on Microorganisms on Frankfurters. J. of Food Science 37:845.

Warnecke, M. D. 1962. Effect of Microbial Flora on Comminuted Meats. M.S. Thesis. The Ohio State University, Columbus, Ohio.

Warnecke, M. D., H. W. Ockerman, H. H. Weiser and V. R. Cahill. 1966. Quality of Processed Comminuted Meat as Affected by Microbial Flora of the Raw Constituents. J. of Food Technology 20:118.

Weiser, H. H., G. J. Mountney and W. A. Gould. 1971. Practical Food Microbiology and Technology. AVI Publishing Co., Inc., Westport, Connecticut.