



History:

Received: February 27, 2018
Reviewed: March 8, 2018
First Published: March 24, 2018
Collection year: 2018
Confirmation of publication: March 24, 2018

Identifiers and Pagination:

Year: 2018
Volume: 3
First Page: 1
Last Page: 7
Publisher Id: AdvFood&NutrSci-3-1
DOI:<http://dx.doi.org/10.21065/>

Corresponding author:

Dr. Nelson A. Cox, Ph.D., Research Microbiologist, Poultry Microbiological Safety and Processing Research Unit, US National Poultry Research Center USDA-Agricultural Research Service, 950 College Station Road Athens, GA 30605.
Phone: 706-546-3484
Fax: 706-546-3633
E: nelson.cox@ars.usda.gov

Citation:

NA Cox, Landrum MA, DE Cosby, ME Berrang, A Hinton Jr., SC Mize, JS Jackson. Reduction of Campylobacter on chicken livers using low acid processing aid. Adv Food & Nutr Sci 3. 1-7

Research Article

REDUCTION OF CAMPYLOBACTER IN CHICKEN LIVERS USING A LOW ACID PROCESSING AID

Landrum MA¹, NA Cox², DE Cosby², ME Berrang², A Hinton Jr.², SC Mize² and JS Jackson¹

1. University of Georgia, Athens, GA;
2. The U.S. National Poultry Research Center, Athens, GA

Abstract

Liver has become a prime source for Campylobacter outbreaks and products are needed to allow processors a more efficient way of controlling foodborne pathogens. Campylobacter reductions in livers treated with a low pH processing aid (CMS PoultryHresh), with and without a surfactant (PoultryHresh Plus) were studied. Chicken livers (n=13/treatment group) were individually inoculated with a C. coli marker strain (10⁷) and each dipped into sterile cups containing 100 mL of water, PoultryHresh or PoultryHresh Plus for 15 s, removed and allowed 5 s to drain. Each liver was placed into 50 mL buffered peptone water and hand shaken for 60 s; controls (n=10) same procedure, no treatment. Rinsates were serially diluted and plated onto Campy Cefex agar with 200 ppm gentamicin. Plates were incubated for 48 h at 42°C microaerobically, colonies counted and log transformed. Procedures were replicated 3 times. Significant reductions in treated compared to untreated for PoultryHresh and PoultryHresh Plus was 98.1% and 99.4%, respectively and with no change in appearance. Treating with this product may allow processors to meet rising performance standards on poultry livers.

Key words: Campylobacter, broiler livers, sanitizer, CMS PoultryHresh

1. Introduction

Campylobacter, the third leading cause of foodborne illness in the U.S., continues to be a major concern to the poultry industry. Illness and/or outbreaks often occur from consumption of raw or undercooked products [1],[2] and may cause serious symptoms including diarrhea, abdominal cramping, fever and vomiting. [3] Poultry products are commonly implicated with outbreaks of campylobacteriosis and investigations worldwide have demonstrated how chicken liver is increasingly becoming a prime source for contamination. [1],[4] In the U.S., Campylobacter was found prevalent in 77% of livers [5], while another study reported Campylobacter in as many as 92.9% of commercial chicken livers. [6] The U.K. experienced a substantial increase in campylobacteriosis associated with liver dishes between 2009 and 2011, causing the Food Standards Agency (FSA) to categorize liver as a high-risk food product. [7] The Centers for Disease Control and Prevention (CDC) investigated and reported serious outbreaks of Campylobacter infections from poultry liver in the U.K. and Australia. [8] In Switzerland, Campylobacter was isolated in livers from 10% to 100%, varying by season [5] and England also reported liver as being a prime source for

Campylobacter outbreaks. [3] Therefore, microbial contamination of broiler livers is a serious, worldwide concern for the industry.

The presence of Campylobacter in chicken liver has become a widespread problem and a serious public health concern. Often cases of campylobacteriosis go undetected and are not reported; therefore, illness and outbreak numbers are probably even higher than predicted, further increasing the seriousness of this foodborne pathogen. [9] Proper procedures for eliminating Campylobacter from livers is thoroughly cooking until an internal temperature exceeding 70 °C is reached for a minimum of 2 minutes. [5], [10],[11] One factor aiding the problem is many liver recipes recommend cooking by 'flash frying', which allows livers to maintain a pink internal color. [5], [11],[12] This cooking method is not adequate to eliminate Campylobacter, allowing it to infect the consumer. [10] This assists in explaining the large number of campylobacteriosis outbreaks in individuals who recently attended catered events or consumed restaurant meals. Research has found that caterers or restaurant cooks are likely to undercook livers in an attempt to maintain the pink coloration consumers desire. [11] This, however, allows many individuals to become sick or an outbreak to occur. Hutchinson et al. [11] described a variety of essential oils and antimicrobial ingredients evaluated as additives for liver recipes, although significant reductions were not found.

Funding:

The authors received no direct funding for this research.

Competing Interests:

The authors declare no competing interests

Additional information is available at the end of the article.

There are some intervention strategies, which include freezing, alternative cooking methods, boiling, chlorinated water, organic acid treatment or pre-cooking treatments. [3], [5], [4],[9],[11] Further research demonstrated Campylobacter to be more prevalent on a liver's outer surface than internally. [13] This indicates external treatment methods may exhibit greater reductions. Past research evaluating organic acid treatment on livers exhibited changes in the surface coloration post treatment, as surface lightening was described as "bleaching". [11] This study investigates the reduction of Campylobacter on livers treated with a low acid processing aid, CMS PoultrypHresh, and the product containing the addition of a surfactant, PoultrypHresh Plus. Differences in past research and this study include the acid used and the length of immersion. Past research used a duration dip time of 2 minutes, whereas the current research is only 15 seconds. [11] If effective, the treatment could potentially prevent cross contamination in a consumer kitchen and lower Campylobacter prevalence internally, as it is not conclusive whether outer contamination seeps into the liver. This treatment may assist processors in reducing contamination levels within processing facilities, as dip time is rapid and potentially a reasonable addition for processing procedures.

2. Materials and Methods

Bacterial Strain

The bacterial Campylobacter strain used for this research is a gentamicin resistant marker strain, *Campylobacter coli* (Cc^{GR}), obtained from Dr. Nelson Cox, USDA, Athens, GA. [14] Initially, Cc^{GR} was streaked onto Campy Cefex Agar [15] containing 200 ppm gentamicin (Sigma, St. Louis, MO). The culture was incubated microaerobically for 48 h at 42°C (5% O₂, 10% CO₂, 85% N₂). Forty-eight-hour (± 4 h) plates of this culture were used to prepare the inocula for this research.

Inoculation of Parts

Livers were obtained from a local grocery (N=36), livers were divided into 3 groups; Trt 1 – low acid processing aide (LAPA) (n=13); Trt 2 – LAPA w/surfactant (LAPAS) (n=13) and Trt 3 – inoculated untreated control (n=10) (Con). A 10^8 suspension of Cc^{GR} (0.1 mL) was used to individually inoculate the surface of each liver. Livers were left undisturbed for 5 minutes to allow the cells an adequate attachment period.

Treatment

Thirteen livers were placed into separate specimen cups containing 100 mL of either PoultrypHresh or PoultrypHresh Plus for 15 s with no agitation. When removed, livers were allowed to drain for 5 s and placed into individual sterile specimen cups containing 50 mL of buffered peptone water. Each liver was hand shaken for 60 s. The controls were inoculated the same as the experimental groups but were not subjected to any treatment before being placed into the specimen cups for rinsing.

Plating and Incubation

After hand rinsing, each rinsate was collected, serially diluted and plated onto Campy Cefex agar with 200 ppm gentamicin. Plates were incubated microaerobically at 42°C for 48 h. Colonies were counted and CFU/mL data was log transformed. All procedures were replicated 3 times.

3. Results

Since Campylobacter in chicken livers is quickly becoming a major concern in the food industry, this study evaluated a potential treatment option to lower prevalence and chance of infection. Results showed the average recovery of C. coli on livers receiving no treatment was 5.5 log₁₀ CFU/mL. After livers were treated with a LAPA 15 s dip, the recovery was reduced to 3.9 CFU/mL. Livers that received a LAPAS 15 s dip were found to have Campylobacter recovery levels lowered to 3.3 log₁₀ CFU/mL. These results indicate a 1.7 log₁₀ reduction (98.1%) when using a LAPA dip and a 2.2 log₁₀ reduction (99.4%) dipping with LAPAS compared to untreated samples (Figure 1). When treated results were compared with a 15 s water dip, LAPA reduced the average log₁₀ CFU/mL by 91.9% (1.1 log₁₀ CFU/mL), while LAPAS lowered C. coli by 97.5% (1.6 log₁₀ CFU/mL). No visible organolytic damage was demonstrated or reported post treatment. Table 1 shows average recovery of C. coli from all replicates, while Table 2 presents the data by replicate.

Table 1. Average Log₁₀ cfu/mL of Campylobacter coli recovered from livers dip treated with no treatment, water, LAPA, or LAPAS for 15 seconds with no agitation (mean±standard deviation)

Treatment	Average Log ₁₀ (cfu/mL)	Reduction from Untreated (%)	Reduction from Water (%)
Untreated	5.5±0.1		
Water	4.9±0.1	76.0	
LAPA	3.8±0.1	98.1	91.9
LAPAS	3.3±0.2	99.4	97.5

Table 2. Average Log₁₀ cfu/mL of *Campylobacter coli* recovered by replicate from livers dip treated with no treatment, water, LAPA, or LAPAS for 15 seconds with no agitation (mean±standard deviation)

Replica te	Treatment	Average Log ₁₀ (cfu/mL)	Reduction from Untreated (%)	Reduction from Water (%)
1	Untreated	5.5±0.3		
	Water	4.9±0.4	73.7	
	LAPA	3.8±0.4	98.0	92.4
	LAPAS	3.1±1.0	99.6	98.5
2	Untreated	5.7±0.2		
	Water	5.1±0.3	74.9	
	LAPA	3.8±0.3	98.8	95.2
	LAPAS	3.6±0.3	99.2	97.0
3	Untreated	5.2±0.2		
	Water	4.5±0.2	79.1	
	LAPA	3.7±0.2	96.9	85.2
	LAPAS	3.1±0.3	99.2	96.2

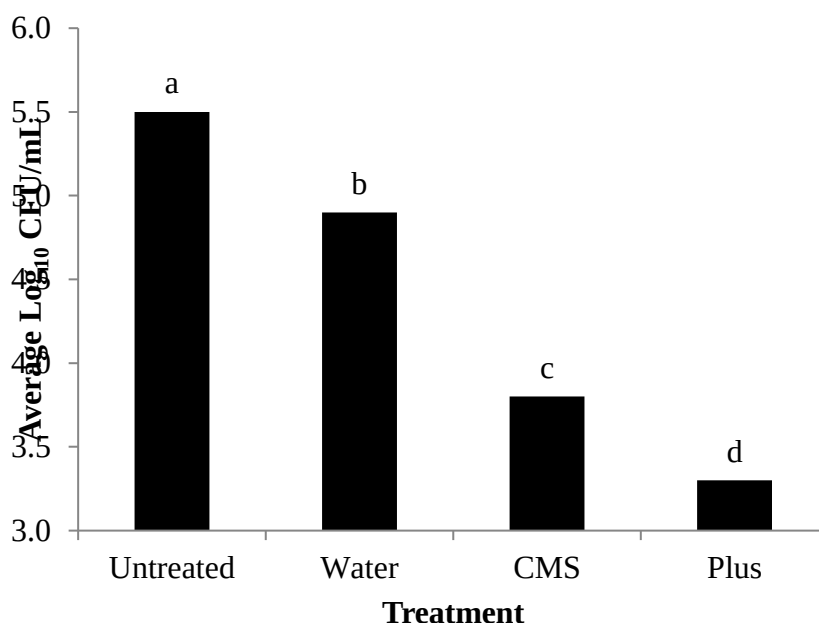


Figure 1. Average log₁₀ cfu/mL of *Campylobacter coli* recovered from livers having been dipped in either no treatment, water, LAPA, or LAPAS for 15 seconds with no agitation.

3. Discussion

A treatment capable of providing such substantial reductions in *Campylobacter* levels is important for poultry producers and consumers worldwide. Alternate treatment options may include the use of chlorinated water, although Bryan and Doyle [9] found that while chlorine may assist in lowering cross contamination between carcasses, it has little effect on bacteria attached to the skin and muscle surfaces. Harrison et al. [5] reported the method of freezing does reduce the presence of *Campylobacter* on the skin and muscle of the broiler. It is likely the consumer may prefer what is considered to be a fresh, never frozen product. Such findings, however, contradict those of Fernandez and Pison [6] who found *Campylobacter* to be highly prevalent in frozen poultry liver.

Additional treatment options include the use of organic acid, which Firlieyanti et al. [4] reported causes color changes/bleaching on the liver surface. Several studies showed it is not effective for internal *Campylobacter* reduction. [4], [11] Noormohamed and Fakhr [3] discussed how foodborne pathogen resistance to antimicrobials is alarming and may arise from cross contamination during processing, possibly causing serious consequences on human health. Their study also demonstrated that the majority of *Campylobacter* isolates were resistant to five of the seven antimicrobials researched and 81 isolates were resistant to more than seven antimicrobials. This further increases the need for a treatment to effectively lower and/or eliminate *Campylobacter* before products are shipped from processing facilities, as Vashin et al. [16] discussed how the likelihood of transferring *Campylobacter* rises in further stages of secondary processing.

Researchers have demonstrated the serious need to reduce *Campylobacter* on poultry liver surfaces throughout the world. Illness and outbreaks arising from livers are increasingly becoming more prevalent and research has found the majority of retail livers are contaminated with *Campylobacter* at varying levels. [4] While the key to *Campylobacter* elimination is allowing adequate cooking times and temperatures, recipes continue suggesting undercooking or flash cooking which could result in illnesses. Intervention strategies within the processing facility are important to lowering *Campylobacter* prevalence and the ample reductions demonstrated in this study may potentially provide the industry with an effective means to reduce the presence of this pathogen and hence human illness. Future research will further evaluate LAPA on a larger scale to reduce *Campylobacter* contamination of poultry products. LAPA may provide another hurdle *Campylobacter* must cross in order to infect consumers effectively reducing the number of campylobacteriosis illnesses associated with poultry products.

References

- [1] Little CL, Gormley FJ, Rawal N, Richardson JF. Short Report: A recipe for disaster: outbreaks of campylobacteriosis associated with poultry liver pâté in England and Wales. *Epidemiol Infect* 2010; 138: 1691-1694.
- [2] Simaluiza RJ, Toledo Z, Ochoa S, Fernandez H. The prevalence and antimicrobial resistance of *Campylobacter* Jejuni and *Campylobacter* coli in chicken livers used for human consumption in Ecuador. *J Anim Vet Adv* 2015; 14: 6-9.

- [3] Noormohamed A, Fakur MK. Incidence and antimicrobial resistance profiling of *Campylobacter* in retail chicken livers and gizzards. *Foodborne Pathog Dis* 2012; 9: 617-624.
- [4] Firlieyanti AS, Connerton PL, Connerton IF. *Campylobacters* and their bacteriophages from chicken liver: The prospect for phage biocontrol. *Int J Food Microbiol* 2016; 237: 121-127.
- [5] Harrison D, Corry JEL, Tchórzewska MA, Morris VK, Hutchison ML. Freezing as an intervention to reduce the numbers of *campylobacters* isolated from chicken livers. *J Appl Microbiol* 2013; 57: 206-213.
- [6] Fernández H, Pisón V. Isolation of thermotolerant species of *Campylobacter* from commercial chicken livers. *Int J Food Microbiol* 1996; 29: 75-80.
- [7] Edwards ES, Milne LM, Morrow K, Sheridan P, Verlander NQ, Mulla R, Richardson JF, Pender A, Lilley M, Reacher M. *Campylobacteriosis* outbreak associated with consumption of undercooked chicken liver pate in the East of England, September 2011: identification of a dose-response risk. *Epidemiol Infect* 2014; 142: 352-357.
- [8] Centers for Disease Control and Prevention (CDC). Multistate outbreak of *Campylobacter jejuni* infections associated with undercooked chicken livers-northeastern United States 2012. *MMWR Morb Mortal Wkly Rep* 2013; 62: 874-876.
- [9] Bryan FL, Doyle MP. Health risks and consequences of *Salmonella* and *Campylobacter jejuni* in raw poultry. *J Food Prot* 1995; 58: 326-344.
- [10] Whyte R, Hudson JA, Graham C. *Campylobacter* in chicken livers and their destruction by pan frying. *Lett Appl Microbiol* 2006; 43: 591-595.
- [11] Hutchison M, Harrison D, Richardson I, Tchórzewska M. A method for the preparation of chicken liver pâté that reliably destroys *Campylobacters*. *Int J Environ Res Public Health* 2015; 12: 4652-4669.
- [12] Strachan NJC, MacRae M, Thomson A, Rotariu O, Ogden ID, Forbes KJ. Source attribution, prevalence and enumeration of *Campylobacter* spp. from retail liver. *Int J Food Microbiol* 2012; 153: 234-236.
- [13] Thompson, T.M., M.E. Berrang, N.A. Cox and R.J. Meinersmann. *Campylobacter* prevalence in retail chicken liver. *Poult Sci* 2018; (Suppl. 1).
- [14] Cox NA, Richardson LJ, Berrang ME, Fedarka-Cray PJ, Buhr RJ. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J Food Prot* 2009; 72: 1288-1292.
- [15] Stern NJ, Wojton B, Kwiatek K. A differential selective medium and dry ice generated atmosphere for recovery of *Campylobacter jejuni*. *J Food Prot* 1992; 55: 514-517.
- [16] Vashin I, Stoyanchev T, Ring Ch, Atanassova V. Prevalence of *Campylobacter* spp. in frozen poultry giblets at Bulgarian retail markets. *TJS* 2009; 7: 55-57.



© 2017 The Author(s). This open access article is distributed under a Creative Commons Attribution (CC-BY) 4.0 license.

You are free to:

Share — copy and redistribute the material in any medium or format

Adapt — remix, transform, and build upon the material for any purpose, even commercially.

The licensor cannot revoke these freedoms as long as you follow the license terms.

Under the following terms:

Attribution — You must give appropriate credit, provide a link to the license, and indicate if changes were made.

You may do so in any reasonable manner, but not in any way that suggests the licensor endorses you or your use.

No additional restrictions

You may not apply legal terms or technological measures that legally restrict others from doing anything the license permits