Meat and Muscle BiologyTM





Decontamination of Chicken Thigh Meat by Pulsed Ultraviolet Light

Joshua R. Cassar, Edward W. Mills*, Jonathan A. Campbell, Ali Demirci

The Pennsylvania State University, University Park, PA 16802, USA *Corresponding author. Email: ewm3@psu.edu (E. W. Mills)

Abstract: Foodborne illness outbreaks associated with chicken can be reduced with effective decontamination interventions. The effectiveness of pulsed ultraviolet (PUV) light for destruction of Salmonella, $E.\ coli$, and Campylobacter on the surface of chicken thigh meat was investigated. Chicken thighs were inoculated to 6 to 7 \log_{10} CFU/cm² before exposure to PUV light for each of the above-mentioned pathogens. Treatment variables included the distance from the quartz window of the PUV light (8 and 13 cm) and treatment time (0, 5, 15, 30, and 45 s). Comparison of treated samples to control (0 s) samples allowed for quantification of microbial reduction due to PUV light treatment. Microbial reduction on lean surface chicken thighs (P < 0.05) with increasing duration of PUV light exposure for $E.\ coli$, Campylobacter, and Campylobacter and Campylobacter

Keywords: *Campylobacter,* chicken thigh, *E. coli,* pulsed ultraviolet light, *Salmonella Meat and Muscle Biology* 3(1):479–487 (2019) **doi:10.22175/mmb2019.08.0033** Submitted 6 Aug. 2019 Accepted 19 Oct. 2019

Introduction

As consumption of poultry products continues to increase, the microbial risks associated with these products increase as well. Between 1988 and 1992, there were 40 foodborne illness outbreaks associated with chicken, accounting for 1.65% of all foodborne illness outbreaks in the United States (CDC, 1996). Between 2009 and 2015, there were 123 chicken associated outbreaks, which accounted for 9.60% of all foodborne illness outbreaks in the United States (Dewey-Mattia et al., 2018). Improving microbial safety of chicken products is important to both consumers and processors (McLeod et al., 2018; Keklik et al., 2010). Salmonella spp., Escherichia coli O157:H7, and Campylobacter are the most prevalent foodborne pathogens associated with raw chicken (USDA, 2012; Haughton et al., 2011; Sarjit and Dykes, 2017; McLeod et al., 2018).

In 2016, the Food Safety Inspection Service (FSIS) within the US Department of Agriculture (USDA) implemented performance standards for *Salmonella* spp. and *Campylobacter* on poultry. This new protocol set the maximum acceptable percent positive for chicken parts at 15.4 and 7.7% for *Salmonella* spp. and *Campylobacter*, respectively. The intention of this protocol is to reduce the occurrence of foodborne illness outbreaks associated with raw poultry (USDA, 2012; Federal Register, 2016). Industry efforts to comply with new performance standards include a variety of antimicrobial interventions applied during chicken processing.

Currently, the poultry industry commonly uses chlorine or organic acid solutions, including peroxyacetic acid, as antimicrobial interventions. Less common are other chemical and physical treatments such as, trisodium phosphate, high hydrostatic pressure, irradiation, pulsed-field electricity, and ultraviolet light (UV; Bolder, 1997; Demirci and Ngadi, 2012; Zhi-Hong et al., 2018). Pulsed ultraviolet (PUV) light is an emerging technology that uses light flashes with a broader spectrum and greater intensity emittance as compared to conventional UV light systems. Conventional UV wavelengths include a spectrum of 100 to 400 nm. The germicidal, UV-C, wavelengths fall between 100 to 280 nm with the maximum germicidal effect at 254 nm. PUV systems use xenon flashlamps to produce a broad wavelength spectrum, 100 to 1,100 nm, with over 50% of the energy deriving from the UV region (Krishnamurthy et al., 2010). Unlike continuous, low intensity output of conventional UV light sources, PUV light is emitted in short bursts of very high intensity light (Bialka and Demirci, 2008; Demirci and Ngadi, 2012; Keklik et al., 2010). In a review by Gómez-López et al. (2007) a continuous UV lamp delivered 0.0039 J/cm² of energy per second and 2 types of xenon pulsed lamps delivered in excess of 0.06 J/cm² per second in the fluence range of 0 to 0.2 J/cm². In 1996, the US Food and Drug Administration (FDA) approved the use of pulsed light (up to 12.0 J/ cm²) for microbial inactivation on foods (FDA, 2015).

Previous research has investigated the application of PUV light as a germicidal intervention for several food products including chicken. Keklik et al. (2010) investigated PUV light's effectiveness in a variety of experiments. In one investigation, inoculated unpackaged and vacuum-packaged boneless/skinless chicken breast samples were treated with PUV light for 5, 15, 30, 45, or 60 s at 5, 8, or 13 cm distance. At 5 cm distance, a 2 log₁₀ CFU/cm² reduction (~99%) was achieved for Salmonella after 15 s for unpackaged samples and at 30 s for vacuum-packaged samples. In the second phase, Keklik et al. (2011) used a pilot-scale PUV light, shackle conveyor system to evaluate the germicidal effect of PUV light on E. coli K12 inoculated on the surface of whole chicken carcasses. The PUV light system in this experiment included a total of four 16-inch lamp housings facing opposite sides of the carcass. Treatment times of 30, 45, 60, 90, 120, and 180 s were selected and corresponded to the time each whole chicken spent in the system. The \log_{10} reductions for this experiment ranged from 0.87 to 1.43 CFU/mL rinse solution after 30 and 180 s treatments, respectively.

The current research is undertaken to investigate the microbial reduction effects of PUV light for boneless chicken thigh, with and without the presence of skin. In this study, the effectiveness of PUV light for inactivation of *Salmonella*, *E. coli*, and *Campylobacter* on the surface of raw lean and skin surface chicken thighs was evaluated.

Materials and Methods

Microorganisms

Salmonella subsp. enterica enterica ser. Typhimurium (ATCC 13311) was obtained from the American Type Culture Collection (Manassas, VA). Escherichia coli K12 was obtained from the E. coli Reference Center at the Pennsylvania State University (University Park, PA). Antibiotic resistant cultures for Salmonella Typhimurium and E. coli K12 were prepared as suggested by Catalano and Knabel (1994), to allow for the use of antibiotic specific media. Nalidixic acid (ARCOS Organics, Geel, Belgium) and streptomycin sulfate (Fisher BioReagents, Fair lawn, NJ) resistant (NSR) cultures of S. ser. Typhimurium and E. coli K12 were obtained from frozen culture stock of Keklik et al., (2010, 2011), respectively. Cultures were maintained in tryptic soy broth (BD-Difco, Franklin Lakes, NJ), supplemented with 0.6% yeast extract and 100 mg/mL of each nalidixic acid and streptomycin sulfate (TSBYE-NS). Salmonella Typhimurium and E. coli K12 were plated on tryptic soy agar (BD-Difco, Franklin Lakes, NJ), supplemented with 0.6% yeast extract and 100 mg/mL of each nalidixic acid and streptomycin sulfate (TSAYE-NS). Colonies of S. ser. Typhimurium NSR and E. coli K12 NSR were both isolated from TSAYE-NS and were used to prepare stock cultures stored in 20% glycerol and kept at -80°C. Working cultures were sub-cultured every 14 d and were maintained in TSBYE-NS at 4°C.

Campylobacter jejuni (ATCC 29428) stock culture was stored in 20% glycerol and kept at -80°C. Campylobacter jejuni stock culture (1 mL) was transferred to 10 mL of Brucella Broth (Criterion, Santa Maria, CA) and incubated at 37°C for 24 to 72 h under anaerobic conditions ($< 0.1\% O_2$ and $> 16\% CO_2$) using a GasPak Anaerobic System (BD, Franklin Lakes, NJ). A loopful of the Brucella Broth was plated on Charcoal Cefoperazone Deoxycholate agar (CCDA-Preston; Oxoid, Hampshire, UK) and incubated at 37°C for 24 to 72 h under anaerobic conditions. Presumptive colonies representing morphology of C. jejuni were confirmed using the Microgen Campylobacter Latex Agglutination Kit (Microgen Bioproducts Ltd., Camberley, UK). Working cultures were sub-cultured every 3 to 5 d and were maintained on CCDA under anaerobic conditions at 37°C. Campylobacter culturing was prepared as described by Scheinberg et al., 2013 with modifications by S. Watson (personal communication, 2017).

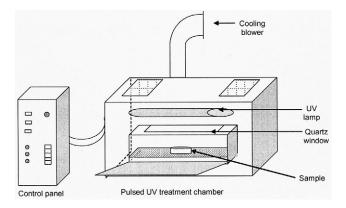


Figure 1. Representation of SteriPulse-XL 3000 Pulsed Ultraviolet Light System (Bialka and Demirci, 2008).

Inoculum preparation

Inoculum preparation was performed as described by Keklik et al. (2010). In separate trials, 1 mL of NSR S. ser. Typhimurium NSR or E. coli K12 NSR working cultures were transferred to 100 mL of TSBYE-NS and incubated at 37°C for 24 h and then centrifuged (10°C at 3,300 × g for 30 min). After incubation the supernatant was removed and sterile 0.1% peptone water (BD, Franklin Lakes, NJ) was used to resuspend the cells. The suspension was centrifuged under the same conditions. After the supernatant was removed, the pellet was resuspended with 50 mL of sterile 0.1% peptone water, yielding 10^8 to 10^9 CFU/mL.

Campylobacter jejuni colonies were isolated on CCDA as previously described, transferred to 10 mL of Brucella Broth and incubated under anaerobic conditions at 37°C for 48 h. After incubation the solution was centrifuged (10° C at $3300 \times g$ for 5 min) and the supernatant was discarded. The pellet was then rinsed with sterile 0.1% peptone water and re-centrifuged under the same conditions as described by Scheinberg et al. (2013). Cells were resuspended with 5 mL of sterile 0.1% peptone water, yielding 10^{6} to 10^{7} CFU/mL.

Chicken thigh meat preparation

Chicken thigh meat was obtained from a local poultry processing plant and kept frozen (ca. -17°C) until use. Chicken meat was transferred to a refrigerator (ca. 2°C) to thaw 48 h prior to each trial. After thawing, thighs were removed from the refrigerator and allowed to warm to ambient temperature (~21°C) 2 h before the trial. Lean or skin surface of chicken thighs were cut to standard sized pieces (5 × 5 × 1 cm).

Cassar et al. Chicken Decontamination by Pulsed UV Light

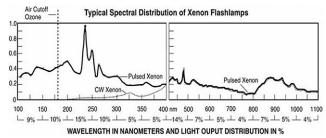


Figure 2. Typical spectral output of a Xenon flashlamp (Sonenshein, 2003).

Inoculation of chicken thigh meat

In individual trials, NSR S. Typhimurium NSR, E. coli K12 NSR, or C. jejuni were applied to the top surface of the chicken thigh samples $(5 \times 5 \times 1 \text{ cm})$ using a micropipette to evenly spread 0.1 mL of the inoculum on the surface area. To obtain 10^5 to 10^6 CFU/cm² on the surface, 0.1 mL of inoculum solution was applied and spread evenly to the top surface of each chicken samples (n = 9). Following inoculation, NSR S. ser. Typhimurium NSR and E. coli K12 NSR samples were held for 1 h at ambient temperature (\sim 21°C) to allow for bacterial attachment. To reduce the death of the microaerophilic C. jejuni, inoculated samples were held for only 30 min at \sim 21°C.

Pulsed ultraviolet light system

Pulsed ultraviolet light was generated using a SteriPulse-XL 3000 Pulsed Light System (Xenon Corporation, Wilmington, MA). The system, represented in Fig. 1, had an energy output of 1.27 J/cm² per pulse at 1.8 cm below the xenon bulb. The xenon bulb pulsed 3 times per second, with each pulse lasting 360 ms. A polychromatic burst was produced with each pulse emitting wavelengths ranging from 100 to 1100 nm. Over 54% of the emitted energy was in the UV spectrum (100 to 400 nm; Sonenshein, 2003; Bialka and Demirci, 2008; Krishnamurthy et al., 2010), graphically depicted in Fig. 2.

Pulsed ultraviolet light treatment

Inoculated thigh samples (n = 9) were individually subjected to PUV light treatment. Treatment variables included the distance from the quartz window of the PUV light unit (8 and 13 cm) and treatment time (5, 15, 30, and 45 s; 3 pulses/s). Nine replications were used for each treatment combination.

Microbial analysis

After treatment with PUV light, chicken samples were transferred to a filtered stomacher bag (Classic 400, Seward Ltd., Worthing, UK) with 25 mL of buffered peptone water (BPW; Oxoid). Samples were then stomached (Model 400, Seward Ltd.) for 2 min at 260 rpm. Solutions filtered out of the samples were serially diluted in 9 mL of BPW. Salmonella ser. Typhimurium NSR and E. coli K12 NSR samples were plated on TSAYE-NS plates using an autoplater (Autoplate 4000, Spiral Biotech; San Diego, CA) and incubated at 37°C for 24 h as described by Keklik et al., (2010). Campylobacter jejuni samples were manually plated since the charcoal-based agar did not allow for enumeration using the auto-count system. Campylobacter jejuni was plated on CCDA plates and incubated at 37°C for 48 h. TSAYE-NS plates were enumerated using an auto-counter (Q-Count, Version 2.1, Spiral Biotech). The CCDA plates were manually counted. For both enumeration methods, counts were expressed as CFU/cm². Log₁₀ reductions were determined by comparing plate counts of treated samples to the plate counts of controls (untreated samples). Salmonella ser. Typhimurium NSR colonies were confirmed using Microgen Salmonella Latex Agglutination Kit (Microgen Bioproducts Ltd., Camberley, UK). Campylobacter jejuni colonies were confirmed using Microgen Campylobacter Latex Agglutination Kit (Microgen Bioproducts Ltd., Camberley, UK). Escherichia coli colonies were visually confirmed by Gram staining.

Surface temperature measurements

In preliminary studies, thigh surface temperature was measured using an infrared thermometer (Lasergrip 800; Etekcity, Anahiem, CA) immediately following PUV treatment. However, this procedure did not allow temperature measurement for product in the chamber during PUV treatment. The observed temperature values were quite variable and declined rapidly while the samples were being removed from the PUV chamber. Thus, it was decided to use a thermocouple inserted immediately beneath the surface as an indication of surface temperature during the PUV treatment.

Surface temperature profile during the PUV light treatment was determined using a type K thermocouple (Omegaette HH306; Omega Engineering Inc, Stamford, CT) with a 15-cm long and 1-mm in diameter thermocouple probe placed approximately 2 mm under the surface of the chicken thigh sample. The sensing area of the thermocouple was the 1×1 mm tip. During exposure to PUV light, temperatures were recorded at 5 s

intervals for up to 45 s at 8 and 13 cm from the quartz window, positioned 5.8 cm below the PUV flashlamp.

Energy measurements

Using a Nova Laser Power/Energy Monitor (P/N 1J06013, OPHIR Optronics Ltd., Wilmington, MA) with a 46-mm aperture pyroelectric metallic absorber (P/N 1Z02860, OPHIR Optronics Ltd.) energy delivered to the location of the sample surface was recorded at 8 and 13 cm from the quartz window, positioned 5.8 cm below the PUV flashlamp. The energy monitor reported total energy delivered. Energy recordings were averaged over 30 pulses and then calculated according to exposure duration to assess total energy (joules/cm²) delivered to the sample.

Statistical analysis

The complete experimental design included 9 replications. Statistical Analysis Software 9.4 (SAS Inst. Inc., Cary, NC) was used as the statistical platform for analysis. An overall 2-way ANOVA with predicator variables proximity and duration was used to assess the microbial reduction associated with proximity, duration and their interaction. When needed, Tukey multiple comparison test was used to establish a significant confidence interval for treatment conditions at 95%. The standard error of the mean (SEM) was provided in tables, when necessary, to represent the deviation of the means among treatments.

Results and Discussion

Log₁₀ reductions due to PUV light exposure for *Campylobacter jejuni*, *E. coli* K12 NSR and *Salmonella* ser. Typhimurium NSR were evaluated and compared for both skin and lean surface chicken thigh. Furthermore, temperature and energy profiles were assessed for each treatment parameter during the PUV light treatment.

Microbial reduction

The log₁₀ reduction of *Campylobacter jejuni*, *Escherichia coli* K12 NSR and *Salmonella* ser. Typhimurium NSR on the surface of raw chicken thighs after treatment with PUV light was assessed at treatment durations of 5, 15, 30, and 45 s and at 8 or 13 cm from the quartz window of the PUV unit. Average treatment durations and proximities resulted in a range of energy fluences delivered to the surface of the product.

Table 1. Effects of PUV light exposure and proximity on \log_{10} reductions of *Campylobacter jejuni*, *E. coli* K12, and *Salmonella* ser. Typhimurium on raw lean surface chicken thighs

		Microorganism		
Parameter		Campylobacter ²	Salmonella ³	E. coli ⁴
Exposure duration, s	5	1.45 ± 0.25^{A}	1.55 ± 0.46^{A}	$1.22\pm0.32^{\rm A}$
	15	$1.69\pm0.23^{\rm B}$	$1.76\pm0.40^{\mathrm{B}}$	1.50 ± 0.39^{AB}
	30	$1.82 \pm 0.17^{\mathrm{B}}$	$1.89\pm0.49^{\rm B}$	1.74 ± 0.23^{BC}
	45	$2.09\pm0.36^{\text{C}}$	2.42 ± 0.67^{B}	2.02 ± 0.23^{C}
Proximity ¹ , cm	8	$1.78 \pm 0.28^{\text{A}}$	$1.88\pm0.59^{\rm A}$	1.63 ± 0.39^{A}
	13	$1.72 \pm 0.38^{\text{A}}$	$1.88\pm0.58^{\text{A}}$	1.61 ± 0.44^{A}

 $^{^{}A-C}$ Means within a column and parameter group without a common superscript are significantly different (P < 0.05).

Lean surface chicken thigh. The distance by treatment time interaction for lean surface samples was not significant (P > 0.05) for microbial reduction of E. coli, Campylobacter or Salmonella. The evaluation of each treatment variable, while holding the other variable constant, showed no significant difference (P >0.05) due to the proximity of the PUV light. Microbial reduction on the surface of chicken thighs after 45 s of PUV light exposure was significantly greater ($P \le$ 0.05) when compared to 5 s of exposure. Lean surface microbial reduction increased (P < 0.05) with PUV light exposure time for E. coli, Campylobacter and Salmonella. Exposure to PUV light for 5, 15, 30, and 45 s on lean surface thighs resulted in log10 reductions of 1.22, 1.50, 1.74, and 2.02 for E. coli, 1.45, 1.69, 1.82, and 2.09 for *Campylobacter*, and 1.55, 1.76, 1.89, and 2.42 for *Salmonella*, respectively (Table 1).

Similar results were reported by Keklik et al., (2010) who studied the effect of PUV light for the reduction of Salmonella ser. Typhimurium on the surface of boneless skinless chicken breast. They reported log₁₀ reductions of Salmonella (CFU/cm²) ranging from 1.2 to 2.4 after a 5 s treatment at 13 cm and a 60 s treatment at 5 cm, respectively. Chun et al., (2010) tested the efficacy of conventional UV at 254 nm for reduction of Campylobacter jejuni, Listeria monocytogenes, and Salmonella ser. Typhimurium. Chicken breast samples were inoculated with 6 to 7 log₁₀ CFU/g and exposed to 0.5 J/cm² of UV (254 nm) radiation. There was a reduction of the populations of C. jejuni, L. monocytogenes, and S. Typhimurium by 1.26, 1.29, and 1.19 log₁₀ CFU/g, respectively. In another study, McLeod et al., (2018) subjected boneless skinless chicken breast

fillets inoculated with pathogenic bacteria to PUV light. Pulsed UV light exposure with fluences ranging from 1.25 to 18 J/cm² resulted in average reductions of 0.9 to 3.0 log₁₀ (CFU/cm²) for Salmonella enteritidis, Listeria monocytogenes, Staphylococcus aureus, Escherichia coli EHEC, Escherichia coli ESBL, Pseudomonas spp., Brochothrix thermospacta and Carnobacterium divergens.

In the current work greater energy fluence (up to 62.24 J/cm²) did not consistently produce greater microbial reduction especially when considering the closer proximity to the PUV light source. Previous reports, generally using lower energy fluence than in this study, show stronger correlation between energy fluence and microbial reduction (Chun et al., 2010; Keklik et al., 2010; McLeod et al., 2018). It seems that some dwell time is needed for development of lethal effects from UV light exposure. Using a pulsed light system allows for very high fluence, but it is not clear that lethality correlates closely with the resulting high UV dose.

Skin surface chicken thigh. The distance by treatment time interaction for lean and skin surface samples was not significant (P > 0.05) for microbial reduction of E. coli, Campylobacter, or Salmonella. The effect of proximity to the quartz window of the PUV light was inconsistent across microbe strains for skin surface thigh. Proximity to the PUV light source had a significant effect on both S. Typhimurium and E. coli with closer proximity resulting in greater reduction (P < 0.05). Distance from the PUV light did not affect skin surface microbial reduction for Campylobacter (P > 0.05). Skin surface microbial reduction increased (P < 0.05) with PUV light exposure time for E. coli, Campylobacter, and Salmonella. PUV light exposure for 5, 15, 30, and 45 s on skin surface thighs resulted in log₁₀ reductions of 1.19, 1.41, 1.66, and 1.96 for E. coli, 1.08, 1.27, 1.54, and 1.85 for Campylobacter, and 0.90, 1.24, 1.58, and 1.82 for Salmonella, respectively (Table 2). A similar study, Haughton et al. (2011) evaluated the reduction of Campylobacter spp., E. coli and S. enteriditis on boneless skinless chicken breast and chicken skin using high intensity light pulses (3 Hz, 505 J/pulse and pulse duration of 360 ms). After 30 s of treatment, inoculated chicken skin had log_{10} (CFU/g) reductions of 1.22, 1.69, and 1.27 for C. jejuni, E. coli and S. enteritidis, respectively. Corresponding reductions on inoculated boneless skinless chicken breast had \log_{10} (CFU/g) reductions of 0.96, 1.13, and 1.35 for C. jejuni, E. coli and S. enteritidis.

¹Distance from the quartz window, which is 5.8cm below the UV strobe.

 $^{^{2}}Campylobacter SEM = 0.047.$

 $^{^{3}}$ Salmonella SEM = 0.072

 $^{^{4}}E.\ coli\ SEM = 0.054$

Table 2. Effects of PUV light exposure and proximity on \log_{10} reductions of *Campylobacter jejuni*, *E. coli* K12, and *Salmonella* ser. Typhimurium on raw skin surface chicken thighs

		Microorganism		
Parameter		Campylobacter ²	Salmonella ³	E. coli ⁴
Exposure duration, s	5	1.08 ± 0.15^{A}	0.90 ± 0.28^{A}	$1.19\pm0.25^{\rm A}$
	15	1.27 ± 0.17^{AB}	1.24 ± 0.29^{B}	1.41 ± 0.30^{AB}
	30	1.54 ± 0.16^{B}	$1.58\pm0.35^{\mathrm{C}}$	1.66 ± 0.39^{BC}
	45	1.85 ± 0.17^{C}	1.82 ± 0.13^{C}	1.96 ± 0.40^{C}
Proximity ¹ , cm	8	$1.43 \pm 0.32^{\text{A}}$	1.52 ± 0.42^{A}	1.62 ± 0.42^{A}
	13	$1.44 \pm 0.34^{\text{A}}$	$1.22\pm0.43^{\rm B}$	$1.47\pm0.38^{\hbox{\footnotesize B}}$

 $^{^{\}mathrm{A-C}}$ Means within a column and parameter group without a common superscript are significantly different (p < 0.05).

Temperature and energy measurements

After warming at room temperature (~22°C) for 2 h, the initial surface temperature of the raw chicken thigh samples was 22.2 ± 2.2 °C. The energy fluence (J/ cm²) and change in temperature were measured under each treatment condition at the sample surface. Values for energy levels and surface temperature are shown in Tables 3 and 4, respectively. The total energy delivered to the chicken samples was expressed as J/cm². Energy levels ranged from 3.4 to 62.2 J/cm² for 5 s at 13 cm and 45 s at 8 cm, respectively. The amount of energy delivered at 8 cm was significantly greater (P <0.05) than the energy delivered at 13 cm. The amount of energy delivered increased significantly (P < 0.05) with increased treatment time at both 8 and 13 cm from the quartz window the PUV light unit. These results are comparable to data reported by Keklik et al., (2010) who also measured total energy of PUV light delivered to boneless skinless chicken breast ranging from 2.9 to 34.8 J/cm² during a 5 s treatment at 13 cm and a 60 s treatment at 5 cm, respectively.

Table 3. Total energy (J/cm²) delivered to the product surface during exposure to pulsed UV light

		Treatme	nt time, s	
Proximity ¹	5	15	30	45
8 cm*	6.92	20.75	41.49	62.24
13 cm**	3.38	10.15	20.30	30.45

 $^{^{\}rm l}{\rm Distance}$ from the quartz window, which is 5.8 cm below the UV xenon flashlamp.

Following PUV treatment chicken thigh surface temperature (average of all treatments) was significantly (P < 0.05) higher for skin surface versus lean surface thigh meat, 33.05°C versus 28.97°C, respectively. The greater temperature rise for the skin surface may be due to greater fat content and lower heat capacity of skin versus lean. Proximity to the PUV light and duration of illumination showed a significant (P < 0.05) interaction for thigh surface temperature, Table 4. Closer proximity and increased time lead to increased temperature with the highest surface temperature observed after 45 s exposure at proximity of 8 cm. Surface temperatures reported in Table 4, from a thermocouple measurement, are somewhat lower than the actual surface temperature from an infrared thermometer. The infrared thermometer readings indicated values of approximately 47.10 and 42.30, respectively, for 8 and 13 cm at 45 s of PUV exposure time while thermocouple readings were 45.98 and 36.40 for comparable samples. In another report for a similar PUV process, temperature increases ranged from 3.9 to 36.8°C after a 5 s treatment at 13 cm and a 60 s treatment at 5 cm, respectively (Keklik et al., 2010). Ozer and Demirci (2006) evaluated the reduction of E. coli and L. monocytogenes on the surface of raw salmon filets after exposure to PUV light. After 60 s of exposure to PUV light, surface temperature increased 91, 68, and 53°C at 3, 5, and 8 cm from the quartz window of the PUV lamp, respectively.

Table 4. Chicken thigh¹ surface temperature (°C) after exposure to pulsed UV light

_					
Proximity ²	0	5	15	30	45
8 cm	22.08 ± 2.19^{A3}	$26.58 \pm 2.24^{\rm B}$	32.10 ± 3.06^{CD}	$39.18 \pm 4.03^{\mathrm{E}}$	45.98 ± 5.51^{F}
13 cm	22.23 ± 2.34^{A}	25.07 ± 2.70^{B}	28.41 ± 3.00^{BC}	$32.40 \pm 3.55^{\text{CD}}$	36.03 ± 3.68^{DE}

A–F Means without the same letter are significantly different (P < 0.05).

¹Distance from the quartz window, which is 5.8cm below the UV strobe.

 $^{^{2}}Campylobacter SEM = 0.047$

 $^{^{3}}$ Salmonella SEM = 0.052

 $^{^{4}}E$. coli SEM = 0.047

^{*}SD for 1 sec of PUV light exposure = 0.014 J/cm^2

^{**}SD for 1 sec of PUV light exposure = 0.025 J/cm^2

¹Average temperature of lean and skin surface thighs.

²Distance from the quartz window.

 $^{^{3}}$ SEM = 1.04.

Table 5. Microbial reduction differs with PUV intensity¹ even when total fluence is similar

Microorganism ²	Lean surface thigh		Skin surface thigh	
	8 cm/15 s	13 cm/30 s	8 cm/15 s	13 cm/30 s
Campylobacter	1.72 ± 0.25	1.83 ± 0.18	1.29 ± 0.17	1.57 ± 0.18
Salmonella	1.58 ± 0.35	1.75 ± 0.46	1.43 ± 0.23	1.44 ± 0.39
E. coli	1.57 ± 0.29	1.76 ± 0.25	1.39 ± 0.29	1.56 ± 0.26
Average of Microorganisms	1.62 ± 0.30	1.78 ± 0.31	1.37 ± 0.23	1.52 ± 0.29
Average (lean vs. skin)	1.70 ± 0.31		1.45 ± 0.27	

¹Intensity is derived from proximity, total fluence is the combination of proximity and duration of exposure. The treatments chosen here were selected for comparison because they share similar total fluence (20.75 J/cm² (8 cm/15 s) and 20.30 J/cm² (13 cm/30 s)).

Discussion

The results of this work demonstrate the effectiveness of PUV light for surface microbial destruction, but also raise questions concerning the importance of surface temperature rise in this process. It was observed that with similar radiant energy fluence skin surface thighs showed greater temperature rise than lean surface thighs. It is speculated that the heat capacity of the skin surface is lower than that of the lean surface. Nevertheless, after treatment of similar fluences, lean surface thighs exhibited greater microbial reduction (Table 5) but less total change in temperature and final temperature (Fig. 3) compared to skin surface thighs. In a study by Murphy et al., (2004), the surface of both skinless and skin-on chicken thighs that were inoculated with Salmonella and Listeria monocytogenes and thermally treated. Skin surface thighs consistently required longer heat treatment with temperatures ranging from 55 to 70°C to achieve a 1 log₁₀ reduction for both Salmonella and L. monocytogenes. Those results suggest that skin surface may offer some protection not provided by lean surface. However, factors measured in the current work cannot explain the apparent contradiction of higher temperature skin resulting in a lower lethality. During long exposure times (>45 s) surface temperatures may reach threshold ranges to allow microbial

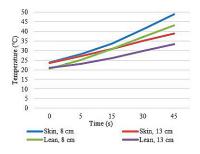


Figure 3. Temperature at the surface of chicken thighs during exposure to PUV light.

growth. However, this is not expected to affect counts in the current study since microbial sampling was completes within minutes following the PUV treatment.

Inversely, with roughly the same energy delivered at 8 cm/15 s (20.75 J/cm^2) and 13 cm/30 s (20.30 J/s^2) cm²), the further proximity and longer duration resulted in greater microbial reduction. The greater microbial reduction at 13 versus 8 cm may be due to the fact that the energy transfer process occurred over a longer duration, 30 versus 15 s. Microbial reduction by thermal treatment is dependent on the rate of rise in the final temperature. The total energy delivered at 8 cm for 15 s was 20.75 J/cm², which is similar to the total energy delivered at 13 cm for 30 s of 20.30 J/cm². Combined results for these 2 treatments showed that lean surface chicken thigh had significantly greater (P < 0.05) germicidal response compared to skin surface thigh. At approximately 20.5 J/cm², lean and skin surface thighs had average microbial reductions of 1.70 and 1.45 log₁₀ CFU/cm², respectively (Table 5). These results could be explained by the fact that skin has a more complex surface than lean muscle. Feather follicles may provide a protective environment for microbes and the dermis layer may accommodate microbial attachment (Cason et al., 2004). A study by Firstenberg-Eden et al. (1978), who evaluated microbe attachment on chicken breast with and without skin reported that skin resulted in greater microbial attachment compared skinless chicken breast. They attributed the increased attachment to differing composition and morphology of the skin. Similar research by Keklik et al. (2010) reported that with increasing energy exposure, and consequent temperature increase, greater microbial reduction is achieved. Clearly, PUV energy fluence and surface temperature must be considered simultaneously. They are not independent predictors of total microbial reduction achievable by PUV light.

 $^{^{2}}SEM = 0.16$

Conclusion

The results from this study demonstrated that PUV light treatment was effective in reducing E. coli, Campylobacter, and Salmonella on the surface of raw chicken thighs. Closer proximity to the PUV lamp, 8 vs. 13 cm, and increased exposure time, up to 45 s, resulted in greater microbial reductions for all pathogens evaluated for both skin and lean surface chicken thighs. Longer treatment times and closer proximity to the PUV light flashlamp resulted in a significant (P < 0.05) increase of surface temperature on the chicken thigh. Ultimately, it is unclear as to whether direct germicidal effects or the increase in temperature contribute more to microbial reduction by PUV light. Also, further research is needed to design a PUV system that reduces shadowing effects associated with treatment of whole carcasses and irregularly shaped parts.

Acknowledgments

This work was supported by the USDA National Institute of Food and Agriculture Federal Appropriations under Project PEN04562. The authors would like to thank Farmers Pride, Bell & Evans (Fredericksburg, PA) for providing chicken samples. Also, the authors would like to thank Xenon Corporation (Wilmington, MA) for the technical assistance on Pulsed UV light system.

Literature Cited

- Bialka, K.L., and A. Demirci. 2008. Efficacy of pulsed UV-light for the decontamination of *Escherichia coli* O157: H7 and *Salmonella* spp. on raspberries and strawberries. J. Food Sci. 73(5):M201–M207. doi:10.1111/j.1750-3841.2008.00743.x
- Bolder, N.M. 1997. Decontamination of meat and poultry carcasses. Trends Food Sci. Technol. 8(7):221–227. doi:10.1016/S0924-2244(97)01040-6
- Cason, J.A., A. Hinton, Jr., and R.J. Buhr. 2004. Impact of feathers and feather follicles on broiler carcass bacteria. Poult. Sci. 83(8):1452–1455. doi:10.1093/ps/83.8.1452
- Catalano, C.R., and S.J. Knabel. 1994. Destruction of *Salmonella enteritidis* by high pH and rapid chilling during simulated commercial egg processing. J. Food Prot. 57(7):592–595. doi:10.4315/0362-028X-57.7.592
- Chun, H.H., J.Y. Kim, B.D. Lee, D.J. Yu, and K.B. Song. 2010. Effect of UV-C irradiation on the inactivation of inoculated pathogens and quality of chicken breasts during storage. Food Control 21(3):276–280. doi:10.1016/j.foodcont.2009.06.006
- Centers for Disease Control and Prevention (CDC). 1996. Surveillance summaries. MMWR 45(No. SS-5). https://www.cdc.gov/mmwr/preview/mmwrhtml/00044241.htm (accessed 12 Sept. 2019)
- Demirci, A., and M.O. Ngadi. 2012. Microbial decontamination in the food industry: Novel methods and applications. Woodhead Publ., Oxford. doi:10.1533/9780857095756

- Dewey-Mattia, D., K. Manikonda, A.J. Hall, M.E. Wise, and S.J. Crowe. 2018. Surveillance for foodborne disease outbreaks—United States, 2009–2015. MMWR Surveill. Summ. 67(No. SS-10):1–11. doi:10.15585/mmwr.ss6710a1
- Federal Register. 2016. New performance standards for *Salmonella* and *Campylobacter* in not-ready-to-eat comminuted chicken and turkey products and raw chicken parts and changes to related agency verification procedures: Response to comments and announcement of implementation schedule. Vol. 81, No. 28. [FR Doc No: 2016-02586]. https://www.fsis.usda.gov/wps/wcm/connect/03910f0e-48f8-4111-ba52-490b07c25c24/2014-0023.htm?MOD=AJPERES (accessed 12 Sept. 2018).
- Firstenberg-Eden, R.U.T.H., S. Notermans, and M. Van Schothorst. 1978. Attachment of certain bacterial strains to chicken and beef meat. J. Food Saf. 1(3):217–228. doi:10.1111/j.1745-4565.1978.tb00275.x
- Food and Drug Administration (FDA). 2015. Code of federal regulations. Pulsed light for the treatment of food: 21CFR179.41.
- Gómez-López, V.M., P. Ragaert, J. Debevere, and F. Devlieghere. 2007. Pulsed light for food decontamination: A review. Trends Food Sci. Technol. 18(9):464–473. doi:10.1016/j. tifs.2007.03.010
- Haughton, P.N., J.G. Lyng, D.J. Morgan, D.A. Cronin, S. Fanning, and P. Whyte. 2011. Efficacy of high-intensity pulsed light for the microbiological decontamination of chicken, associated packaging, and contact surfaces. Foodborne Pathog. Dis. 8(1):109–117. doi:10.1089/fpd.2010.0640
- Keklik, N.M., A. Demirci, and V.M. Puri. 2010. Decontamination of unpackaged and vacuum-packaged boneless chicken breast with pulsed ultraviolet light. Poult. Sci. 89(3):570–581. doi:10.3382/ps.2008-00476
- Keklik, N.M., A. Demirci, and R.G. Bock. 2011. Surface decontamination of whole chicken carcasses using a pilotscale pulsed UV light system. Trans. ASABE 54:993–1000. doi:10.13031/2013.37083
- Krishnamurthy, K., J.C. Tewari, J. Irudayaraj, and A. Demirci. 2010. Microscopic and spectroscopic evaluation of inactivation of *Staphylococcus aureus* by pulsed UV light and infrared heating. Food Bioprocess Technol. 3:93–104. doi:10.1007/s11947-008-0084-8
- McLeod, A., K. Hovde Liland, J.E. Haugen, O. Sørheim, K.S. Myhrer, and A.L. Holck. 2018. Chicken fillets subjected to UV-C and pulsed UV light: Reduction of pathogenic and spoilage bacteria, and changes in sensory quality. J. Food Saf. 38(1):e12421. doi:10.1111/jfs.12421
- Murphy, R.Y., T. Osaili, L.K. Duncan, and J.A. Marcy. 2004. Thermal inactivation of *Salmonella* and *Listeria*. monocytogenes in ground chicken thigh/leg meat and skin. Poult. Sci. 83(7):1218–1225. doi:10.1093/ps/83.7.1218
- Ozer, N.P., and A. Demirci. 2006. Inactivation of *Escherichia coli* O157: H7 and *Listeria* monocytogenes inoculated on raw salmon fillets by pulsed UV-light treatment. Int. J. Food Sci. Technol. 41(4):354–360. doi:10.1111/j.1365-2621.2005.01071.x
- Sarjit, A., and G.A. Dykes. 2017. Transfer of *Campylobacter* and *Salmonella* from poultry meat onto poultry preparation surfaces. J. Food Prot. 80(5):750–757. doi:10.4315/0362-028X. JFP-16-414

- Scheinberg, J., S. Doores, and C.N. Cutter. 2013. A microbiological comparison of poultry products obtained from farmers' markets and supermarkets in Pennsylvania. J. Food Saf. 33(3):259–264. doi:10.1111/jfs.12047
- Sonenshein, A.L. (2003). Killing of *Bacillus* spores by high-intensity ultraviolet light. Sterilization and decontamination using high energy light. Xenon Corporation, Wilmington, MA.
- United States Department of Agriculture (USDA). 2012. Food Safety and Inspection Service. Office of Public Health Science Microbiology Division, the nationwide microbiological baseline data collection program: Raw chicken parts survey, January 2012–August 2012. https://www.fsis.usda.gov/wps/wcm/connect/a9837fc8-0109-4041-bd0c-729924a79201/Baseline_Data_Raw_Chicken_Parts.pdf?MOD=AJPERES (accessed 10 Sept. 2018).
- Zhi-Hong, Z., W. Lang-Hong, Z. Xin-An, H. Zhong, and B.S. Charles. 2018. Non-thermal technologies and its current and future application in the food industry: A review. Int. J. Food Sci. Technol. doi:10.1111/ijfs.13903