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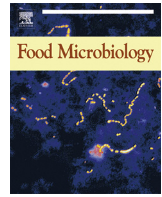

Short communication

Influence of in-package cold plasma treatment on microbiological shelf life and appearance of fresh chicken breast fillets

Jiamei Wang [a, b], Hong Zhuang [c], Arthur Hinton Jr. [c], Jianhao Zhang [b, *]

[a] College of Food Science and Technology, Hainan University, Haikou, 570228, China
[b] National Center of Meat Quality & Safety Control, Nanjing Agricultural University, Nanjing, 210095, China
[c] Agricultural Research Service, USDA, Athens, GA, 30605, USA

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ABSTRACT

The effect of in-package cold plasmas (CP) was studied on microbiological shelf life and surface lightness of fresh chicken fillets (pectoralis major). Chicken fillets were packaged in food trays in air or modified atmosphere (MA) gas (O_2:CO_2:N_2 = 65:30:5) and stored at 4 °C after exposed to an in-package cold plasma (80 kV for 180 s) treatment. Populations of mesophiles, psychrophiles, and *pseudomonas* spp. were measured as indicators for microbiological shelf life and CIELAB L^* values as an indicator for raw meat appearance. Results show that regardless of microbial type, there were no significant differences in microbial counts between the control and CP treated chicken fillets packed in air. However, in the MA packages, microbial counts were consistently lower than the non-treated control during refrigerated storage. Regardless of CP treatment, the microbial counts on the samples packed in air were much higher than in MA. They were more than 6 logs cfu/g in air compared to fewer than 4 logs cfu/g in MA after 7 d storage and fewer than 6 logs cfu/g after 14 d storage. Regardless of CP treatment and gas composition in package, there were no significant differences in the surface L^* value between the fillets pre-treatment and those after storage at 4 °C. These results demonstrate that the effects of in-package CP treatments on microbiological shelf life of fresh chicken fillets depend upon headspace composition in packages. When fresh chicken fillets are packed in air, CP treatment has no effect on microbiological shelf life. MA packages with high O_2 and CO_2 significantly extend shelf life and CP treatment with MA can at least double shelf life of fresh chicken meat (more than 14 days). Regardless of headspace composition, in-package CP does not have negative effects on chicken meat appearance.

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1. Introduction

Contamination of fresh chicken meat with spoilage bacteria has been a challenge for poultry industry. Each year, it is estimated that more than 144 million pounds of fresh poultry meat products are lost as a result of microbiological spoilage in the US (Russell, 2009).

Cold plasma (CP) is an emerging non-thermal treatment and decontaminates meat products effectively in the experiments. Rod et al (Rod et al., 2012) reported that a 20 s CP treatment of a ready-to-eat meat product bresaola resulted in reduced *Listeria innocua* concentrations by 0.8–1.6 log cfu/g. Fröhling et al (Fröhling et al., 2012) found that after a CP treatment, total aerobic plate counts from fresh pork meat remained between 10^2 and 10^3 cfu/g during a 20-d storage period at 5 °C. Noriega et al (Noriega et al., 2011) used CP to treat chicken meat and chicken skin contaminated with *L. innocua* and found that under optimal conditions an 8-min treatment resulted in a 1 log *L. innocua* reduction on skin, while a 4-min treatment produced a >3 log reduction on muscle. With bacon slice, Kim et al (Kim et al., 2011) found that the decrease of pathogen bacteria was 1–2 logs after helium CP treatment, while it was 2–3 logs after helium/oxygen CP treatment.

In-package CP is one of CP generation methods and involves the use of a dielectric barrier discharge (DBD) to generate cold plasma (Misra et al., 2014a,c). In DBD, CP is generated between two electrodes at high potential difference, separated by one or more dielectric barriers. When the potential across the gap reaches the breakdown voltage, the dielectric acts as a stabilizing material leading to the formation of a large number of micro-discharges (Misra et al., 2014a,c, 2013). The in-package CP system allows treatment over large volumes in air and discharge gaps, when

* Corresponding author. College of Food Science and Technology, Nanjing Agricultural University, 1# Weigang, Nanjing, 210095, China.
E-mail address: nau_zjh@njau.edu.cn (J. Zhang).

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sufficiently high potential difference is maintained across the gas gap, and allows treatment of produce inside sealed packages, which eliminates the risk of post-process contamination. Recently several experiments were conducted to evaluate the in-package CP for extending shelf life of fresh produce. Misra et al. (2014b) treat fresh strawberries with the in-package CP and reported that the background microflora (aerobic mesophilic bacteria, yeast and mould) of strawberries was reduced by 2 log within 24 h of post-ACP treatment and effect of CP on product color and firmness was insignificant. Ziuzina et al. (Ziuzina et al., 2014) treated cherry tomatoes with the in-package CP and found that the CP treatment for 10, 60 and 120 s resulted in reduction of *Salmonella*, *Escherichia coli* and *Listeria monocytogenes* populations on tomato to undetectable levels from initial populations of 3.1, 6.3, and 6.7 log CFU/sample, respectively. Further study (Misra et al., 2014a) showed that there were no significant differences among weight loss, pH, and firmness between control and CP-treated cherry tomatoes at the end of storage life.

Although those findings demonstrate that the in-package CP treatments can be used effectively to reduce microbial populations and extend shelf life of fresh produce, there is lack of systematic study on the in-package CP for extension of shelf life of chicken meat products. The ultimate goal of our research was to develop an in-package CP treatment for extending shelf life of packaged fresh chicken meat. The specific objective of the present study was to evaluate the in-package CP effects on microbial populations and appearance of treated chicken breast meat during refrigerated storage.

2. Material and methods

2.1. Cold plasma system

A DBD plasma system was developed based on previous work and designs used to treat spinach leaves and *Bacillus subtilis* spore strips (Keener et al., 2012; Klockow and Keener, 2009). The system consisted of an AC Dielectric Test Set (BK-130, Phenix Technologies, Accident, MD, USA), high voltage wires, aluminum electrodes (150 mm diameter), and dielectric barriers (polypropylene sheets). Chicken meat samples were packaged in sealed trays in air or modified atmosphere (MA).

2.2. Sample packaging

Fresh chicken breast fillets (pectoralis major) were collected from a local broiler processing plant. Samples were transported to lab (ARS-USDA, Athens, GA) on ice in a cooler in 20 min.

Two fillets (93.5 ± 2.5 g each) were weighed and placed in a Cryovac CS977 polymeric tray (Cryovac Sealed Air Corp., Duncan, SC, USA). One fillet was used for microbial analysis and the other for surface (skin side) lightness (CIELAB L* value) measurements. The trays were filled with either ambient air or MA gas (65% O_2, 30% CO_2, 5% N_2) according to the experimental design using a gas mix (Gas Mixer KM-Flow, Witt-Gasetechnik, Deutschland, Germany) and sealed by a tray sealer (Koch Kats 100 Single Head Tray Sealer, UltraSource LLC, Kansas City, Missouri, USA). For the MA package, the gas compositions in package were verified with a headspace gas analyzer (CheckPoint-Handheld Gas Analyzer, Dansensor, DK-4100 Ringsted, Denmark) and the actual headspace compositions were 63.7 ± 2.8% O_2 and 29.2 ± 2.0% CO_2. After sealing, samples were placed on a refrigerated cooler for more than 1.5 h to let relative humidity reach more than 80% in packages (the relative humidity was determined using EL-USB-2-LCD + RH/Temp data logger [Lascar Electronics Inc. UK] sealed in the package) before CP treatments.

2.3. In-package plasma treatment

The packaged trays with meat samples in center were placed directly between electrodes (a direct treatment) and treated with DBD at 80 kV for 180 s at a 5.2-cm electrode gap. Treatments were performed at ambient air and temperature (25 ± 2 °C) and atmospheric pressure. Controls were packaged in the same way and remained on the counter for 3 min. Ozone formation in packages was used as an indicator for CP formation and measured with Drägger tubes (Drägger, Germany) immediately after treatment, because ozone formation has been the most commonly used indicator for in-package DBD-based CP treatment due to its relatively longer half life and relatively easily and quantitatively measured (Klockow and Keener, 2009; Misra et al., 2013, 2014c,b; Ziuzina et al., 2014). Four treatments were included and they are 1) air control (AC); 2) air + CP treatment (AT); 3) MA control (MAC); and 4) MA + CP treatment (MAT).

2.4. Shelf life

Treated samples were stored at 4 °C until the specified sampling days. On days 0, 3, 7, 10, and 14, three packages from each treatment were randomly removed from the cold room. From each package, one fillet was used for microbial measurements and the other for surface color analysis.

2.5. Surface color measurement

The surface color (L^*) of chicken fillets was measured by using a spectrophotometer (CM-700-D, KONICA MINOLTA SENSING, INC., Japan) with settings of illuminant C, 10° observer, specular component included, and an 8-mm aperture on the same fillets before they were sealed in the package and after the packages were opened to avoid the variation in lightness of chicken fillets. Surface areas were selected that were free from obvious defects (bruises, discolorations, hemorrhages or any other conditions that might have prevented uniform color readings). Three measurements were taken at different surface locations for each fillet.

2.6. Microbial analysis

The chicken fillet from each package was place in a sterile bag with 25 ml sterile 1× PBS solution and the bag was then shaken for 1 min. The PBS solution was then diluted by 1:10 for series times in sterile 1× PBS solution before 100 μL of each dilution was taken and spread on TSA plates (Becton Dickinson, Franklin Lakes, NJ, USA) for mesophiles or psychrophiles. For mesophiles, the plates were incubated at 37 °C for 24 h. For psychrophile, they were incubated at 4 °C for 10 days. *Pseudomonas* agar (Becton Dickinson, Franklin Lakes, NJ, USA) was used for determining population of *Pseudomonas*. The plates were incubated at ambient temperature (25 ± 2 °C) for 2 days. Number of microbial colonies was counted and expressed as log cfu/g fresh fillet weight.

2.7. Statistical analysis

All data reported were the means of three different trials with the standard errors. The statistical analysis was performed with one-way analysis of variance, and significant differences between mean values were identified by Duncan's multiple range test using SAS 8.2 software (SAS institute Inc., Cary, NC, USA) with a confidence level of $P < 0.05$.

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