TREATMENT TIME AND VOLTAGE EFFECTS OF DIELECTRIC BARRIER DISCHARGE ATMOSPHERIC COLD PLASMA TO REDUCE *E.COLI* O157:H7 ON PORK *LONGISSIMUS LUMBORUM*

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I.   INTRODUCTION

Food spoilage and food poisoning are mainly caused by microorganisms, which usually exist in processed as well as unprocessed foods [1]. The dielectric barrier discharge (DBD) atmospheric cold plasma is a novel non-thermal sterilization method which has a number of industrial applications [2]. DBD parameters, i.e. treatment time and voltage, differently affect the spoilage bacteria on the fresh meat. *Escherichia coli* O157:H7, a toxin-producing food and waterborne bacterial pathogen, has been linked to large outbreaks of gastrointestinal illness for more than two decades [3]. However, there is a lack of reports on the treatment time and voltage effects of DBD atmospheric cold plasma to reduce *E.Coli* O157:H7 on pork lion. The objective of the present study was to investigate the inactivation of *E.Coli* O157:H7 under different treatment time and voltage of DBD atmospheric cold plasma on pork lion.

II.   MATERIALS AND METHODS

Sample preparation and inoculation

Fresh, raw, pork *longissimus lumborum* (LL) was purchased at a local supermarket in Nanjing province and cut into 2 cm thickness steaks of about 100 g and associated external subcutaneous fat was trimmed off under sterile conditions.

E.coli O157:H7 (NCTC 12900) employed for this experimental work was obtained from the National Collection of Type Cultures (NCTC, Beijing, China). *E.coli* O157:H7 was cultivated in Modified EC broth containing 20ppm of Novobiocin at 37 °C for 24 h and harvested by centrifugation, washed and re-suspended in saline solution (0.85% NaCl, 10^8 CFU/ml) before being used for the superficial contamination in pork LL muscle. The culture suspension (0.1 ml) was inoculated and spread on the prepared pork loins. The pork loins placed in the polypropylene tray and sealed with oxygen barrier film based on polyamide / polyethylene. To facilitate attachment of the *E.coli* O157:H7 to the samples, the samples were incubated for 2 h.

Treatment of dielectric barrier discharge plasma

Dielectric Barrier Discharge System used in this research was the same as that described by Wang *et al.* [4]. The trays with samples were placed in center of electrodes and directly treated between two electrodes (4.5 cm gap) at 60, 75 and 90 kV for 1,3,5,7 min, respectively.

Microbial analysis

After treatment, samples were stored at 4 °C for 2 h. Then, samples were blended with sterile saline for 2 min using a stomacher (BagMixer 400, Interscience Ind., St. Nom, France). A series of decimal dilutions was prepared with sterile saline. Media used for the enumeration Viable cell counts was Sorbitol Maconkey Agar Base (Hopebiol Co., Ltd., Beijing, China). The plates were incubated at 37 °C for 48 h, and then the numbers of colonies on the plates were counted.

Data analysis

All data reported were the means of three different trials with the standard errors, Statistical analysis was carried out with the statistical analysis system (Version 9.0, SAS Institute, Cary, NC, USA, 2006). Statistical analysis was performed by one-way analysis of variance (ANOVA). Significant differences among samples was determined by least square difference method at a confidence level of $P < 0.05$.

III.   RESULTS AND DISCUSSION

To assess the effects of DBD plasma against *E.Coli* O157:H7 on the surface of meats, we treated the samples with DBD plasma for a range of times between 1 and 7 min under different voltages. *E.Coli* O157:H7 were determined after DBD plasma at different exposure conditions as shown Fig. 1. The survival curves of DBD plasma treatment at 60 kV with respect to the treatment time were one-line curves and at condition of 75 and 90 kV were biphasic curves on the surface of meats (Fig. 1), Exposure time required to inactivate 90% of *E.Coli* O157:H7 on the surface of meats(D values) at the condition of 60, 75 and 90 kV were 17.8, 8.3 and 6.02 min. Meantime, difference of sterilization effect of DBD plasma against *E.Coli* O157:H7 on the surface of meats was significant between 60 kV and 75 or 90 kV. Similar research has been reported that one-line curves for the inactivation of *Listeria monocytogenes* in sliced cheese at 75 and 100 W and survival curves change to biphasic curves at 125 and 150 W [5].
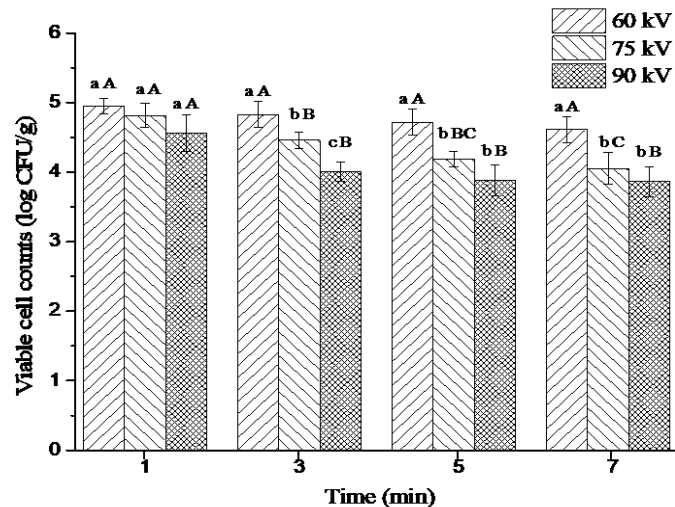

Figure 1. Treatment time and voltage effects of DBD to reduce *E.Coli* O157:H7 in pork LL muscle

IV.   CONCLUSION

In conclusion, our findings showed that DBD plasma successfully decreased the levels of *E.Coli* O157:H7 on the surface of meats. Viable cell counts of *E.Coli* O157:H7 decreased as the DBD plasma treatment time increased. Moreover, the results of this study indicated that in order to obtain higher efficiency of sterilization by DBD plasma on the surface of meats, higher voltage may be required. Therefore, DBD plasma treatment can be applicability to protective of food on survival of microorganism.

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